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Complement Receptors in Inflammation

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1. Introduction

Complement was first discovered in 1889 as a bactericidal protein, distinct from heat stable antibodies present in normal serum. Since that time, it has been shown that the complement system is a biochemical cascade, comprised of more than 30 fluid phase and membrane-associated proteins, normally present as inactive forms. The complement system can be activated by different sequential cascades of enzymatic reactions (described below) in which proteins are sequentially cleaved and activated. The resulting effector molecules, C3a and C5a (also known as anaphylatoxins) are the most potent complement activation products, showing diverse activities on many cell types ranging from chemoattraction to apoptosis. The main target cells carry specialized complement receptors through which anaphylatoxins participate in host defense, inflammatory processes, and immune responses.

The complement system plays an essential role in innate immunity by defending the host against bacterial, viral, and parasitic invasion. Complement proteins promote opsonisation and/or phagocytosis and intracellular killing of these pathogens by immune effector cells such as macrophage and neutrophils. Complement proteins, particularly those of the classical pathway, aid in the processing of immune complexes and in protection against the development of immune complex diseases such as systemic lupus erythematosus (SLE). Recently, it has become evident that the complement system also regulates adaptive immunity involving B and T cells that help in the elimination of pathogens. Furthermore, the engagement of complement receptors on antigen-presenting cells (APC) and other immune cells leads to production of immunoregulatory cytokines. Not only is complement involved in innate and adaptive immunity, it is also involved in pathological conditions. For example, in allergic disease complement proteins participate in the development of an inflammatory reaction. The complement pathways are also activated in patients with sepsis, allergic rhinitis, allergic asthma, and allergic skin conditions such as urticaria. This chapter will outline how complement proteins, C3a and C5a, and their complement receptors regulate inflammation.

2.1.2 Lectin pathway

The lectin pathway is initiated when PRRs, MBL, H-, M- and L-ficolin, recognize and bind sugar moieties on yeast, bacteria, parasites and viruses. In the circulation, these PRRs associate with MBL-associated serine protease 1 (MASP1), MASP2 and MASP3, and a truncated MASP2 known as MAP19. Binding of the MBL-MASP complex to pathogen results in the cleavage of C4 and C2 and the generation of a C3 convertase (C4bC2a; Figure 1), similar to that of the classical pathway (Wagner and Frank, 2010).

2.1.3 Alternative pathway

The alternative pathway is triggered by carbohydrates, lipids and proteins found on pathogen. Slow and constant hydrolysis of circulating C3 (C3 tickover) and its interaction with complement factors B, and D leads to formation of a C3 convertase (C3bBb; Figure 1) (Wagner and Frank, 2010).

2.1.4 Extrinsic protease pathway

In addition to the above three pathways, a C3 independent pathway can also activate the complement system. An extrinsic protease pathway involves direct cleavage of C3 and C5 by a series of proteolytic enzymes released by neutrophils and macrophages; and factors such as Kallikrein, plasmin and factor XIIa. Thrombin, a coagulation factor, can directly cleave C5 to generate biologically active C5a in C3-deficient mice in which C5 convertase cannot be formed (Ricklin et al., 2010).

2.2 Complement proteins C3a, C4a and C5a

The complement proteins, C3a, C4a and C5a, are components of the complement system that are produced from C3, C4 and C5, respectively, as part of the complement activation cascade. C3a is a 9 kiloDalton (kDa) peptide fragment released during selective proteolytic cleavage of the C3 α chain by a C3 convertase of the classical or the alternative pathway. C4a is a 8.7 kDa peptide released from the α chain of C4 by C2a cleavage in an early step of the classical pathway. C5a is an 11 kDa peptide released from the α chain of C5 by action of either classical or alternative pathway C5 convertase (Ember and Hugli, 1997).

Although C3a, C4a and C5a are genetically related, they have marked structural differences in their sequences. Only 13 residues are conserved between C3a, C4a and C5a molecules from several species examined. Six of the conserved residues are cysteines whose side chains participate in forming three intrachain disulfide linkages that stabilize the folded alpha-helical peptide chain. The three disulfide bonds in C3a are Cys 22-Cys 49, Cys 23-Cys 56 and Cys 36-Cys 57. The four-helix bundle of C5a is stabilized by Cys 21-Cys 47, Cys 22-Cys 54 and Cys 34-Cys 55 disulfide bonds. Further structural similarities include a flexible carboxyl terminal tail -L-G-L-A-R in C3a, -A/V-G/H-L-A/Q-R in C4a, and -M/I/V-Q-L-G-R in C5a, which forms a helical turn connected by a short loop, which is important for effector functions of these proteins (Ember and Hugli, 1997).

C3a, C4a and C5a are potent inflammatory peptides with diverse activities on many cell types. They act as chemoattractants for immune cells such as neutrophils, eosinophils, mast cells, and monocytes recruiting to sites of injury or inflammation. They also regulate vasodilation, increase the permeability of blood vessels, and induce smooth muscle contraction. Anaphylatoxin stimulation can induce oxidative burst in neutrophils, histamine release from mast cells and basophils, production of eosinophil cationic protein (ECP) from

eosinophils, and production of pro-inflammatory cytokines from monocytes, B cells and T cells. Characterization of the inflammatory activities of complement proteins indicate relative activities in the order of C5a>C3a>C4a on most tissues examined (Hugli, 1981).

The complement proteins generated as a result of the complement activation damage host tissues. It is therefore obvious that control mechanisms are needed to tightly regulate these potent peptides and maintain homeostatic balance. Indeed, once C3a and C5a are cleaved from C3 and C5 respectively, they are rapidly degraded by plasma enzyme carboxypeptidases. The resulting C3a desArg lacks any pro-inflammatory activity; however, C5a desArg exhibits a reduced inflammatory activity of 1-10% compared with C5a. Interestingly, C3a desArg, also known as acylation stimulating protein (ASP), has been described as possessing metabolic hormone activity that drives triglyceride synthesis and glucose uptake. This suggests a regulatory role of C3a desArg in cell apoptosis and lipid metabolism (Cianflone et al., 1989).

3. Complement receptors for C3a and C5a: C3aR, C5aR and C5L2

The complement proteins C3a and C5a exert their pleiotropic effects by binding to a family of three receptors belonging to the G protein-coupled receptor (GPCR) superfamily. These receptors are the C3a receptor (C3aR), the C5a receptor (C5aR) and the C5a receptor-like 2 (C5L2).

3.1 C3a receptor (C3aR)

C3aR is a membrane glycoprotein of approximately 54 kDa (Ames et al., 1996). This receptor displays high affinity for C3a with a dissociation constant (K_d) of about 1 nM, but not for C3a desArg or C5a (Crass et al., 1996). Human platelets express a high molecular weight (95-105 kDa) variant of C3aR that binds C3a with K_d of 8×10^{-10} M.

3.1.1 Structure

As a GPCR, C3aR contains seven transmembrane domains within its 482 amino acids sequence. C3aR distinctively possesses a large second extracellular loop between the fourth and fifth transmembrane domain that is indispensable for ligand binding. This loop contains 175 amino acid residues; in most GPCR the corresponding extracellular loop is 30-40 amino acids long. Sulfation of tyrosine 174 in this loop is essential for binding C3a (Chao et al., 1999). The genes encoding C3aR have been mapped to p13.2-3 of chromosome 12 in humans and 6F1 in mouse (Hollmann et al., 1998). C3aR displays 50-60% homology between various species, with 65% sequence identity between human and murine counterparts (Hollmann et al., 1998).

3.1.2 Expression

C3aR is expressed on cells of myeloid origin, including monocytes/macrophages, neutrophils, eosinophils, basophils, mast cells, dendritic cells and microglia. Additionally, C3aR is also expressed on non-myeloid cells, such as astrocytes, endothelial cells, epithelial cells, smooth muscle cells, and activated T cells. One paper has described the receptor's expression on human tonsillar B cells; while others have confirmed the absence of C3aR on human B cells. The receptor is also expressed in tissues from lung, liver, kidney, brain, heart, muscle and, testis.

3.1.3 Biological role

C3aR mediates chemotaxis of eosinophils (Daffern et al., 1995), mast cells (Nilsson et al., 1996), dendritic cells (Gutzmer et al., 2004) and monocytes, but not neutrophils (Daffern et al., 1995). It can also trigger oxidative burst in macrophages, neutrophils and eosinophils (Burg et al., 1996). In addition, basophils (Bischoff et al., 1990) and mast cells (Johnson et al., 1975, Venkatesha et al., 2005) undergo degranulation and release histamine upon C3a-C3aR interaction. C3aR stimulates production of ECP from eosinophils (Takafuji et al., 1994), as well as upregulation of β_2 -integrins and shedding of L-selectins, thereby promoting eosinophil adhesion to endothelial and epithelial cells (Jagels et al., 2000). Human monocytes and mast cells exhibit increased intracellular calcium (Ca^{2+}) levels when stimulated with C3a (Venkatesha et al., 2005). C3a also stimulates smooth muscle contraction (Stimler et al., 1983), lysozyme release from immune cells (Showell et al., 1982), platelet aggregation (Becker et al., 1978), and triglyceride synthesis in adipocytes (Baldo et al., 1993).

3.1.4 Signaling

Upon C3a binding to the C3aR, intracellular signal transduction is promoted via heterotrimeric guanosine triphosphate (GTP)-binding proteins (G proteins). C3aR mediates its effect on immune cells via coupling to the pertussis toxin (PTX)-sensitive and -insensitive G proteins $\text{G}\alpha_i$ and $\text{G}\alpha_{16}$, respectively. In endothelial cells, C3aR also couples to PTX-insensitive $\text{G}\alpha_{12}$ and $\text{G}\alpha_{13}$ (Schraufstatter et al., 2002). Downstream signaling events involve activation of phosphoinositol-3-kinase gamma (PI3K- γ), which in turn activates phospholipase C (PLC) β and PLC γ . This leads to generation of inositol triphosphate (IP3) and diacylglycerol (DAG), leading to Ca^{2+} mobilization and phosphokinase C (PKC) activation, respectively. PI3K can also activate Raf/mitogen-activated protein kinase (MAPK)/extracellular-signal regulated kinase (ERK) kinase (MEK)/ERK cascade inducing expression of various pro-inflammatory cytokines and chemokines.

3.2 C5a receptor (C5aR or CD88)

First cloned in 1991 (Boulay et al., 1991, Gerard and Gerard, 1991), C5aR is a membrane glycoprotein of approximately 42 kDa (Richardson et al., 1998), displaying high affinity for C5a and C5a desArg. Human C5aR binds C5a with a K_d of 1 nM, and with 10 to 100-fold lower affinity to C5a desArg (K_d of 412-660 nM) whereas C3a and C3a desArg are not recognized (Monk et al., 2007).

3.2.1 Structure

Similar to C3aR, C5aR is a seven transmembrane GPCR that belongs to the rhodopsin-like family. Based on the analysis of transmembrane domain sequences, C5aR is clustered with other chemoattractant receptors, such as C3aR, formyl peptide receptor family, ChemR23, platelet activating factor (PAF) receptor, IL-8 receptor, and bradykinin receptor (Methner et al., 1997, Samson et al., 1998). The C5aR gene is localized to q13.3-13.4 of human chromosome 19 (Gerard et al., 1998). Murine C5aR exhibits 65% sequence identity with its human counterpart (Gerard et al., 1992). The N terminus of C5aR is required for high affinity binding of C5a, but not for receptor activation (DeMartino et al., 1994, Mery and Boulay, 1993). A second distinct binding site is formed by charged residues in the second and third extracellular loops and the external faces of the transmembrane helical bundle and

hydrophobic residues in the core of the C5aR. Unlike the N terminal binding site, the second site is responsible for receptor activation (Gerber et al., 2001). Current evidence suggests that at least three different discontinuous regions exist within the C5a molecule for interaction with C5aR (Huber-Lang et al., 2003).

3.2.2 Expression

C5aR is expressed on cells of myeloid origin such as neutrophils, eosinophils, basophils, mast cells, dendritic cells and monocytes, as well as on non-myeloid cells, including bronchial and alveolar epithelial cells, endothelial cells, Kupffer cells, stellate cells, astrocytes and microglial cells (Monk et al., 2007).

3.2.3 Biological role

C5aR is a powerful chemoattractant receptor for monocytes (Pieters et al., 1995), neutrophils (Webster et al., 1980), eosinophils (DiScipio et al., 1999), basophils (Lett-Brown and Leonard, 1977), mast cells (Hartmann et al., 1997), B cells (Kupp et al., 1991) and T cells (Nataf et al., 1999). It also stimulates mast cell degranulation (Subramanian et al., 2011), mast cell chemotaxis in specific mast cell subtypes (Hartmann et al., 1997, McCloskey et al., 1999), oxidative burst in granulocytes and the production of reactive oxygen species (ROS) in neutrophils (Guo et al., 2003), secretion of lysosomal enzymes from macrophages (McCarthy and Henson, 1979) and polymorphonuclear (PMN) cells as well as the secretion of pro-inflammatory mediators from monocytes, eosinophils (Takafuji et al., 1994) and mast cells (Hartmann et al., 1997). C5a is also responsible for upregulation of vascular adhesion molecules such as E-selectin, inter-cellular adhesion molecule 1 (ICAM-1) and vascular cell adhesion molecule 1 (VCAM-1) after systemic activation of complement (Albrecht et al., 2004, Jagels et al., 2000). Our preliminary findings suggest that C5a can promote human mast cell adhesion to extracellular matrix protein, as well as human mast cell migration *in vitro*, indicating a critical role of C5a/C5aR in inflammation.

3.2.4 Signaling

C5aR signaling depends on heterotrimeric G proteins. C5aR mainly couples to $G\alpha_{i2}$, a PTX-sensitive G protein (Skokowa et al., 2005). However, ectopically expressed C5aR, and C5aR on cells of the hematopoietic lineage, can also couple to PTX-insensitive $G\alpha_{i6}$ (Amatruda et al., 1993, Monk and Partridge, 1993). C5a-C5aR interaction leads to activation of several components of signaling pathway, including PI3K- γ , PLC β 2, and phospholipase D (PLD) (Rabiet et al., 2007). C5aR can activate the transcription factor, cyclic AMP (cAMP) response element-binding protein (CREB), by phosphorylation at the convergence of two pathways, PI3K/Akt and ERK (Perianayagam et al., 2006). CREB activation has been proposed to be a part of the mechanism by which C5a can delay neutrophil apoptosis and prolong an inflammatory response (Perianayagam et al., 2004). In neutrophils, C5a causes downstream activation of p21-activated kinases (PAK), which are downstream effectors of cdc42 and rac (small guanosine triphosphate (GTP)-binding proteins (GTPases)), as well as $G\gamma$ subunits (Huang et al., 1998). PAK family members are involved in altering cell morphology/chemotaxis, activation or potentiation of several distinct MAPK cascades and the activation of nuclear factor (NF)- κ B in macrophages. p38 MAPK is activated by PAK1/PAK2 and, in turn, activates MAPK-activated protein kinase 2 (MAPKAP-K2). In primary macrophages from MAPKAP-K2-deficient mice, chemotaxis to C5a is impaired.

Furthermore, the p38 MAPK inhibitor, SB203580, can inhibit C5a-induced migration in a mouse acute lung injury model (Rousseau et al., 2006).

C5aR also couples directly or indirectly to a small range of other intracellular proteins. The Wiskot-Aldrich syndrome protein (WASP) was detected as a binding partner of the C-terminus of C5aR using a yeast two-hybrid assay. WASP binding was strongly potentiated in the presence of active cdc42, suggesting that the association occurs after C5aR activation (Tardif et al., 2003). WASP is a multifunctional protein with a role in the regulation of actin dynamics, and so could be involved in the chemotactic response to C5a. In human erythroleukaemia cells, signal transducers and activators of transcription (STAT3) phosphorylation can be stimulated by C5a in a PTX-insensitive manner, most likely through $G\alpha_{16}$ and the Ras/Raf/MEK/ERK and janus kinase (JAK) pathways (Lo et al., 2006). In contrast, STAT3 phosphorylation occurs only through an ERK pathway in C5a-stimulated neutrophils (Kuroki and O'Flaherty, 1999).

3.3 C5a receptor-like 2 (C5L2)

First discovered in 2000 as a putative orphan receptor (GPR77), (Ohno et al., 2000) C5L2 has since been identified as a second C5a receptor. It is a 37 kDa protein and binds C5a with high affinity (K_d of 2.5 nM). Unlike C5aR, C5L2 binds C5a desArg with a 20-30 fold higher affinity (Cain and Monk, 2002).

3.3.1 Structure

C5L2 consists of 337 amino acids with asparagine 3 as a potential glycosylation site (Ohno et al., 2000). In the conserved transmembrane regions, C5L2 shares 58% sequence identity with C5aR and 55% with C3aR (Lee et al., 2001). Unlike C5aR, C5L2 uses critical residues in its N terminal domain for binding only to C5a desArg. In addition to C5a and C5a desArg binding, C5L2 has been considered a binding partner for C3a, C3a desArg (Kalant et al., 2005), C4a and C4a desArg; however the available data is not very convincing (Johswich et al., 2006, Okinaga et al., 2003).

Although C5L2 has the conventional structure of a GPCR, studies have found that C5L2 does not couple to G proteins. This may be due to a structural difference in the third transmembrane domain. In GPCR, a highly conserved DRY motif in the third transmembrane domain is important for its interaction with the corresponding G proteins. The DRY motif is DRC in C3aR and DRF in C5aR, but DLC in C5L2. Moreover, the third transmembrane domain of C5L2 is truncated. Mutation of DLC in human C5L2 to DRC has been shown to increase coupling to $G\alpha_{16}$ in co-transfected human embryonic kidney (HEK)293 cells and induce a functional response (Okinaga et al., 2003). Interestingly, no functional response occurs in rat basophilic leukemia cells (RBL-2H3) using a C5L2-mutant where the DRY-motif and two additional regions typically involved in G protein coupling are replaced by the corresponding C5aR sequences (Scola et al., 2009). Taken together, these findings strongly suggest that C5L2 completely lacks the potential to couple with G proteins.

3.3.2 Expression

C5L2 is expressed in various tissues of myeloid and non-myeloid origin and transcripts are detected in brain, placenta, ovary, testis, spleen and colon. Surface expression of C5L2 has been detected in lung, liver, heart, kidney, adipose tissue, skin fibroblasts, neutrophils, and

immature, but not mature dendritic cells. In the presence of a C5aR antagonist, binding of C5a has been demonstrated on differentiated myeloblastic HL-60, U937, and epithelial HeLa-cells. C5L2 and C5aR seem to be frequently co-expressed in most cells or tissues (Monk et al., 2007).

3.3.3 Biological role and signaling

C5L2 is an enigmatic receptor as available data suggest opposing roles. On the one hand, C5L2 has been described as a non-signaling decoy receptor for C5a and C5a desArg. In contrast, some studies suggest that C5L2 serves as a signaling functional receptor. In support of a role as a decoy receptor, no mobilization of intracellular Ca^{2+} occurs in C5L2 transfected cells after C5a administration (Cain and Monk, 2002, Okinaga et al., 2003). Moreover, no Ca^{2+} mobilization occurs in neutrophils from C5aR-deficient mice after stimulation with C5a (Hopken et al., 1996) or in C5L2 expressing epithelial and myeloid cell lines (Johswich et al., 2006). In the presence of C5a and C5a desArg, C5L2 transfected RBL-2H3 cells do not degranulate (Cain and Monk, 2002). Further, C5a binding to C5L2 in bone marrow cells derived from C5aR-deficient mice fails to induce any changes in messenger RNA (mRNA) expression (Okinaga et al., 2003).

Nevertheless, there is accumulating evidence that C5L2 is a functional receptor capable of regulating C5aR function *in vitro* and *in vivo*. Neutrophils and macrophages from C5L2-deficient mice produce more tumor necrosis factor (TNF) α and interleukin (IL)6 in response to stimulation with C5a and lipopolysaccharide (LPS) than their wildtype (WT) counterparts. Further, C5a+LPS stimulation drive more IL-6 production from rat neutrophils when C5L2 is blocked (Gao et al., 2005). *In vivo*, C5L2-deficient mice suffer from augmented inflammatory responses and higher numbers of infiltrating neutrophils in a model of pulmonary immune complex injury (Gerard et al., 2005), indicating an anti-inflammatory function of C5L2. The anti-inflammatory role of C5L2 is further supported by a study in which LPS-injected C5L2-deficient mice show higher IL-1 β levels and decreased survival rates (Chen et al., 2007). Similarly, C5L2-deficient mice display higher serum concentrations of IL-6 as compared to WT and C5aR-deficient mice in a model of septic peritonitis (Rittirsch et al., 2008).

In contrast, under the same conditions, a strong reduction of other inflammatory mediators, such as IL-1 β , macrophage inflammatory protein (MIP)-1 α and MIP-2 is observed in C5L2-deficient mice compared to WT mice. Indeed, the plasma concentrations of these mediators are comparable to those found in C5aR-deficient mice. Furthermore, C5L2-deficient mice, like C5aR-deficient mice, or mice in which either of the receptors are blocked by anti-receptor antibodies, show a higher survival rate in mid-grade sepsis (Rittirsch et al., 2008). Contrasting the *in vitro* findings that C5a+LPS-driven IL-6 production by mouse neutrophils is increased when C5L2 is blocked by antibodies (see above), Chen et al. found that IL-6 release is reduced from C5L2-deficient neutrophils (Chen et al., 2007). Moreover, C5a+LPS-induced Mac-1 surface expression on these neutrophils also diminishes, suggesting neutrophils chemotaxis may be impacted. Likewise, whereas C5a or C5a+LPS stimulation can lead to strong ERK1/2- and AKT-phosphorylation in neutrophils from WT mice, there is only a weak effect in the absence of C5L2. Additionally, C5L2-deficient macrophages have an impaired induction of co-stimulatory molecules (CD40, CD86). Even C3a mediated effects such as ERK1/2- and Akt-phosphorylation or F-actin formation on neutrophils are impaired in C5L2-deficient mice (Chen et al., 2007). In addition to these *in vitro* findings,

inflammatory responses are reduced *in vivo* in models of thioglycollate induced peritonitis, thioglycollate induced migration into dorsal air pouches, and ovalbumin (OVA) induced airway hyperresponsiveness (AHR) (Chen et al., 2007). Thus, these studies point to a more complex role of C5L2 in inflammation with C5L2 acting not only as a decoy receptor but also as positive modulator of C5aR and even C3aR. Although C3a desArg does not bind directly to C5L2, overexpression of C5L2 or its downregulation by antisense oligonucleotides influences the effects of C3a desArg (Kalant et al., 2005), suggesting that C5L2 can modulate signaling pathways of other receptors.

Although C5L2 does not appear to signal using the traditional mechanisms employed by GPCRs, several studies suggest that C5L2 has the ability to induce cellular effects. A recent study has found that noradrenaline upregulates C5L2 message and protein in rat astrocytes, and this correlates with an anti-inflammatory response induced by noradrenaline. Transfection of astrocytes by C5L2 down regulates NF- κ B activity, whereas antisense oligonucleotides against C5L2 induce the reverse effect (Gavrilyuk et al., 2005). These observations suggest that the presence of C5L2 may exert some inhibitory effects within the cell, although the mechanisms behind such responses are currently unknown.

4. Regulation of complement receptor signaling

GPCR represent the largest family of cell surface receptors in the human genome, allowing extracellular signals to regulate a vast number of physiological events. Following ligand binding, GPCR undergo a conformational change which activates heterotrimeric G proteins. G proteins exist as a complex of $G\alpha$ and $G\beta\gamma$ subunits. Upon activation, GDP is displaced by GTP from the $G\alpha$ subunit resulting in disassociation of $G\alpha$ subunit from the $G\beta\gamma$ subunits. This facilitates the free subunits interactions with various effector molecules and initiates downstream signaling (Pundir and Kulka, 2010). In the immune system, GPCRs play a role in innate, adaptive and pathological responses. In allergic diseases, stimulation of the GPCRs, C3aR and C5aR, by C3a and C5a, respectively produces a diverse array of effector functions, ranging from inflammatory cell migration to pro-inflammatory mediator production, thus contributing towards the pathophysiology of the diseases.

Given that the activation of C3aR and C5aR can induce massive inflammation and tissue destruction, there are mechanisms in place to limit complement activation where and when it occurs. As mentioned before, after generation, C3a and C5a are quickly degraded by plasma carboxypeptidases. The enzyme cleave the C-terminal arginine, resulting in C3a desArg and C5a desArg formation, each having less than 10% of their original biological activity (Bokisch and Muller-Eberhard, 1970). Formation of C3a and C5a is also regulated by either preventing the assembly of C3 convertase or, once it is formed, by inhibiting its activity. This is accomplished through the actions of decay acceleration factor (DAF), C4 binding protein (C4BP) and Factor H (Sarma and Ward, 2011). In addition, desensitization to prolonged or repeated exposure to high agonist concentration is another important mechanism of GPCR regulation. Furthermore, G proteins may terminate their own activation by $G\alpha$ hydrolysis of GTP, thereby allowing $G\alpha$ -GDP to reunite with $G\beta\gamma$ and form an inactive heterotrimer (Tsang et al., 1998). As this reaction proceeds at a slow rate (Tsang et al., 1998), additional cofactors come into play that aid in GPCR desensitization. Indeed, there are around 30 isoforms of Regulator of G protein signaling (RGS) proteins, which bind to activated $G\alpha$ subunit and accelerate their intrinsic GTPase activity (Willars,

2006). RGS13 can regulate human mast cell lines, HMC-1 and laboratory of allergic diseases (LAD)2, response to C5a. RGS13-deficient HMC-1 cells have more cytosolic Ca^{2+} in response to C5a, indicating that RGS13 regulates C5aR-stimulated events in immune cells (Bansal et al., 2008).

GPCR desensitization is also achieved by the G protein-coupled receptor kinase (GRK)-arrestin pathway. There are seven GRKs in humans, GRK1-7, and four arrestins, arrestin 1-4. Most GPCRs are regulated by only four GRKs: GRKs 2, 3, 5, or 6, and two arrestins: arrestin-2 (β -arrestin1) and arrestin-3 (β -arrestin2) (Premont and Gainetdinov, 2007). Agonist-induced desensitization of GPCRs occurs via a multistep process. Just as G proteins recognize activated GPCR, GRKs also recognize activated GPCR, which leads to receptor phosphorylation at various serine/threonine residues on the intracellular loops and the carboxyl-terminal tail (Langkabel et al., 1999). Upon phosphorylation by GRK, GPCR's affinity for arrestin proteins is increased, which prevents the receptor from activating additional G proteins. The β -arrestins interact with clathrin and the adaptor protein complex AP-2 and target the agonist occupied receptors to pre-existing clathrin-coated pits for internalization. Thus GRK phosphorylation and arrestin binding result in termination of GPCR signaling, despite the continued presence of agonist (Santini et al., 2002, Scott et al., 2002).

A study by Ahamed et al. has shown that C3a stimulates degranulation and chemokine monocyte chemoattractant protein-1 (MCP-1) production in RBL-2H3 cells expressing C3aR. Stimulation with C3a causes phosphorylation of WT C3aR but not of phosphorylation-deficient C3aR (Δ ST-C3aR). In addition, C3a stimulation increases degranulation only in RBL-2H3 cells expressing Δ ST-C3aR, suggesting that receptor phosphorylation provides an "off" signal for degranulation. Overexpression of GRK2 in C3aR-transfected cells results in increased C3a-induced C3aR phosphorylation. This increase in receptor phosphorylation is associated with a significant inhibition of degranulation. Furthermore, C3a causes a rapid translocation of β -arrestin2 from the cytosol to the membrane in RBL-2H3 cells expressing C3aR but not in cells expressing Δ ST-C3aR, indicating that GRK2-induced C3aR phosphorylation is required for β -arrestin recruitment following C3a stimulation (Ahamed et al., 2001). Similarly, in transfected COS cells, overexpression of GRK2, GRK3, GRK5 or GRK6 results in enhanced C3a-induced C3aR phosphorylation (Langkabel et al., 1999). Knockdown of GRK2 or GRK3 expression in HMC-1 and LAD2 cells, causes higher Ca^{2+} mobilization, attenuated C3aR desensitization, and enhanced degranulation, thus indicating GRK2 and GRK3 involvement in C3aR desensitization. On the other hand, GRK5 or GRK6 knockdown in HMC-1 and LAD2 cells has no effect on C3aR desensitization, but instead promotes C3a-mediated degranulation, suggesting a complex role for GRKs in regulating human mast cells (Guo et al., 2011).

A recent study has identified the roles of β -arrestin1 and β -arrestin2 on C3aR desensitization and internalization (Vibhuti et al., 2011). By stably knocking down their expression in HMC-1 cells, this study shows that β -arrestin2, but not β -arrestin1, is required for C3aR desensitization and internalization. Interestingly, β -arrestin1-deficient, but not β -arrestin2-deficient, HMC-1 cells show reduced NF- κ B activation and chemokine MIP-1 β generation in response to C3a. Similar knock-down study with LAD2 cells shows that the absence of β -arrestin1, but not β -arrestin2, significantly inhibits C3a-induced degranulation. This demonstrates that β -arrestin1 and β -arrestin2 play a distinct role on C3aR desensitization, internalization and mediator generation in human mast cells (Vibhuti et al., 2011).

Upon ligand binding, C5aR undergoes rapid phosphorylation of the six serine residues located in the carboxyl-terminal tail (Giannini et al., 1995). This phosphorylation is a critical step in the termination of the C5aR signaling since a phosphorylation-deficient C5aR mutant triggers sustained intracellular signaling leading to increased C5a-induced superoxide production by HL-60 cells (Christophe et al., 2000). A study by Braun et al. shows that a persistent complex between activated/phosphorylated C5aR and β -arrestins is necessary for receptor desensitization and internalization. WT C5aR and β -arrestin1-transfected RINm5F cells exhibit significant β -arrestin1 recruitment to the plasma membrane when stimulated with C5a. Moreover, in resting RINm5F cells, the C5aR is mainly located in the plasma membrane, and both β -arrestin1 and β -arrestin2 are evenly distributed throughout the cytoplasm. Upon addition of C5a, β -arrestin1 and β -arrestin2 rapidly move from the cytosol to the plasma membrane, co-localize with C5aR, promote endocytosis of C5aR and remain associated to C5aR-containing vesicles. Furthermore, this study also indicates that C5aR is mainly internalized via the classical clathrin-dependent pathway (Braun et al., 2003). Interestingly, an observation by Bamberg et al. reveals that in resting neutrophils C5L2 is distributed throughout the cytoplasm co-localized with β -arrestin, while C5aR is expressed on their surface. Following stimulation with C5a, association of both C5aR and C5L2 with β -arrestin is greatly increased. Cells treated with an anti-C5aR antibody when activated with C5a fail to translocate C5L2 to β -arrestin, indicating that the activation of C5L2 is a consequence of C5aR activation. Moreover, C5L2- β -arrestin complex is a negative regulator of C5aR signaling in neutrophils (Bamberg et al., 2010).

Thus, a number of mechanisms have evolved to efficiently regulate complement activation; however, these regulatory mechanisms seem to fail in various clinical and experimental situations underlining the important role of complement proteins and their complement receptors in the pathogenesis of inflammatory diseases. These include airway inflammation and asthma (Wagner et al., 1999), sepsis (Gao et al., 2005), multiple organ dysfunction (Mastellos et al., 2005), hyperacute graft rejection (Link et al., 1999), ischemic injuries of various organs (Arumugam et al., 2004), autoimmune disorders such as SLE (Karp, 2005) or multiple sclerosis (Rus et al., 2005), neurological diseases such as stroke (Yanamadala and Friedlander), and cancer (Yan et al., 2008).

5. Diseases associated with complement function

5.1 Allergic inflammation: Allergic asthma

Asthma is a chronic inflammatory disorder of the airways characterized by variable airflow obstruction, and associated increase in airway responsiveness to a variety of stimuli. The clinical features of asthma include dyspnea, wheezing and coughing. It is thought to be mediated primarily by allergen-specific CD4⁺ T cells, T helper (Th)2 cytokines, and allergen-specific IgE, leading to pulmonary inflammation and AHR. Complement is well recognized as an important factor in the pathophysiology of asthma.

In allergic asthma and allergic rhinitis conditions, the complement system is activated by several pathways leading to the cleavage of C3 and/or C5 into their active fragments. First, the presence of preexisting allergen-specific antibodies in asthmatic patients can activate the classical pathway through formation of immune complexes. Second, the alternative pathway can be initiated by nucleophilic attack of C3 directly on the surface of allergen or by Factor B (Taube et al., 2006). Third, the recognition of PAMPs and danger-associated molecular patterns (DAMPs), such as nucleic acids on apoptotic cells (Elward et al., 2005) or

allergen polysaccharide, (Bito, 1977, Zimmermann et al., 1989) can activate the lectin pathway. Fourth, proteases released from inflammatory cells (Huber-Lang et al., 2002) or direct protease activity of allergens (Maruo et al., 1997) can drive the generation of C3a and C5a. Indeed, a variety of allergens have been shown to activate the complement and generate C3a and C5a in the airways. Ragweed extracts activate the complement cascade in both allergic individuals and healthy controls; however, generation of C3a is much stronger in allergic patients (Gonczi et al., 1997, Hidvegi et al., 1995). Ragweed allergen challenge also promotes C3a generation in the nasal mucosa of patients with allergic rhinitis. House dust mite (HDM), *Aspergillus fumigatus* and perennial ryegrass extracts induce serum C3a and C5a production in a dose and time-dependent manner (Nagata and Glovsky, 1987). Proteases from *Dermatophagoides* spp. (in particular, Der p 3 and Der f 3) can cleave C3 and C5 into their active fragments (Castro et al., 1991, Maruo et al., 1997). During an ongoing allergic reaction the Th2 cytokine IL-4 induces C3 mRNA production in human and murine epithelial cells (Khirwadkar et al., 1993).

Collectively, these studies provide strong evidence that major complement activation, and C3a and C5a generation occurs in asthmatic individuals. C3a and C5a are important contributing factors in the pathophysiology of asthma, because of their ability to recruit and activate inflammatory immune cells such as mast cells, eosinophils, macrophages, neutrophils, and basophils, increase vascular permeability, stimulate smooth muscle contraction, and trigger mast cell degranulation (Wust et al., 2006). Several studies have reported marked anaphylatoxins production under asthmatic condition. In the serum of asthmatic patients, an increase in C3a and C5a levels have been observed after allergen-induced bronchospasm (Arroyave et al., 1977, Smith et al., 1990). The levels of C3a, and to a lesser extent C5a increase in the bronchoalveolar lavage fluid (BALF) of asthmatic patients after segmental allergen provocation. Similarly, allergen-challenged lung lobes show significantly higher C3a levels in comparison to diluent (sham)-challenged lobes from asthmatic individuals (Humbles et al., 2000). In addition, sputum from patients with asthma exhibit increased C5a/C5a desArg concentrations (Marc et al., 2004). Infact, plasma C5a desArg levels reflect allergic disease severity (Bowser et al., 2010). Complement C3a and C4a concentrations increase in plasma of patients with aspirin-induced asthma (Lee et al., 2006). Studies with animal models of AHR have also indicated that C3a and C5a are crucial for asthma pathogenesis (Abe et al., 2001, Bautsch et al., 2000, Drouin et al., 2002, Lukacs et al., 2001).

In addition to allergen-mediated mechanisms, environmental stimuli can also trigger complement activation. Airborne pollutants such as diesel exhaust particles or airborne particulate matter can activate complement through the alternative pathway in human serum and airway epithelium, respectively (Walters et al., 2002). Moreover, acute ozone exposure can induce AHR and neutrophil infiltration, accompanied by elevated C3 levels in BALF in mice (Lambrecht and Hammad, 2009, Park et al., 2004). Cigarette smoke has been shown to directly activate the alternative pathway through cleavage of the internal thioester bond in C3 (Kew et al., 1987). Consistent with these studies, elevated levels of C3 have been found in children from smoking homes compared to those from non-smoking homes (Shima and Adachi, 1996).

Candidate gene and genome-wide screens for asthma susceptibility loci have identified C5 (9q34) (Ober et al., 1998, Wjst et al., 1999) and C5aR (19q.13.3) gene-containing chromosomal regions to be linked to asthma susceptibility (CSGA, 1997, Ober et al., 1998). One study reports an association between a single nucleotide polymorphism (SNP) in the C3 gene and

atopic asthma in children and adults in a Japanese population. On the other hand, SNPs in the human *C5* gene are associated with protection against both childhood and adult asthma. The authors also report a significant association between a SNP in the *C3a* receptor (*C3ar1*) gene and childhood asthma (Hasegawa et al., 2004). These observations may explain the opposing role of *C3* and *C5* in animal-based asthma studies. *C3*-deficient mice when challenged with an allergen to induce pulmonary allergy exhibit diminished AHR and lung eosinophilia. Furthermore, these mice show markedly reduced IL-4 secreting cells and attenuated allergen-specific IgE response (Drouin et al., 2002). Collectively, these results demonstrate that *C3* promotes Th2 effector function in asthma. On the other hand, the A/J mouse strain in which allergic inflammation and AHR can be easily induced, are deficient in *C5* (Karp et al., 2000), demonstrating that *C5*-deficient mice are more susceptible to experimental asthma. Furthermore, *in vitro* experiments have shown that peritoneal macrophages from *C5*-deficient A/J mice produce significantly less IL-12, because IL-12 is critical for promotion of Th1 immune response (Karp et al., 2000), these studies suggest that dysfunction in *C5* cleavage may influence susceptibility to asthma. In contrast, a study by Peng et al. has demonstrated the contribution of *C5* in the development of airway inflammation, AHR and ongoing allergic response in OVA-sensitized mice (Peng et al., 2005). Pharmacological inhibition of *C5* using a monoclonal antibody against *C5*, BB5.1, markedly attenuates airway obstruction, perivascular and peribronchial infiltration of inflammatory cells, and BALF levels of inflammatory mediators such as eotaxin, regulated upon activation, normal T-cell expressed and secreted (RANTES), TNF α , and matrix metalloproteinase (MMP)9 (Peng et al., 2005).

The paradoxical effects of *C5* and *C5a* observed in allergic asthma may be due to *C5* behaving differently during allergen sensitization as opposed to during established allergic inflammation. Evidence to this effect was provided by Kohl et al. in a study utilizing two models of pulmonary asthma: a) OVA sensitization and challenge of mice leading to inhalation tolerance; and b) HDM sensitization and challenge of mice which induce Th2 sensitization, airway inflammation, and AHR. Ablation of *C5aR* signaling by pharmacological targeting prior to initial pulmonary OVA challenge, using a neutralizing anti-*C5aR* monoclonal antibody, induces significant production of Th2 cytokines (IL-5, IL-13, and IL-10), high serum IgE levels, marked influx of inflammatory cells (eosinophils, neutrophils, and lymphocytes) into BALF, mucus production, and AHR. Similar results are observed when *C5aR* is blocked prior to initial pulmonary HDM challenge. Together, this data suggests that the presence of *C5a* in the airways at the time of initial allergen encounter serves to prevent the development of Th2-mediated immune response (Kohl et al., 2006).

Protection against allergic sensitization by *C5aR* activation is a complex process involving alterations in the function of APCs, in particular dendritic cells. Dendritic cells strategically located in the upper layers of the epithelium and lamina propria of the airways, are specialized for the recognition and internalization of PAMPs. Once loaded with allergen, dendritic cells migrate to draining lymph nodes and present antigen to naïve T cells, which under appropriate polarizing cytokine signals, differentiate into effector T cells (Banchereau and Steinman, 1998). Dendritic cells are composed of several subsets, some of which drive the development of an aberrant adaptive immune response. The two dendritic cell subsets which have been found in the bronchial airways are myeloid dendritic cells (mDCs) and plasmacytoid dendritic cells (pDCs) (GeurtsvanKessel and Lambrecht, 2008). Pulmonary mDCs function as the major APCs in mouse models of allergic asthma, preferentially

inducing Th2 immune response towards inhaled allergen (Lambrecht et al., 2000). Pulmonary pDCs are immature dendritic cells, which induce respiratory tolerance by directly suppressing mDC-mediated activation of CD4⁺ T effector cells or by inducing regulatory T cells (T_{regs}, a heterogeneous population of T cells that can suppress allergic immune responses) (GeurtsvanKessel and Lambrecht, 2008).

Several studies suggest that the ratio between mDCs and pDCs is critical for the development of the dysregulated Th cell response and that C5a regulates this ratio, thereby modulating the development of the allergic phenotype during allergen sensitization. CD4⁺ T cells from mice challenged with HDM allergen in presence of an anti-C5aR monoclonal antibody produce higher levels of Th2 cytokines in co-culture with mDCs, but not with pDCs. Importantly, in the same setting, pDCs suppress the mDC-induced production of Th2 cytokines (Kohl et al., 2006). Furthermore, exposure to HDM leads to an increased number of mDCs, whereas the number of pDCs remains unaffected. These findings demonstrate that C5aR signaling controls the accumulation of pulmonary mDCs and pDCs in lungs, keeping the mDC:pDC ratio low, which facilitates the suppressive effects of pDCs on the mDC-mediated activation of T cells. In addition, C5aR blockade positively regulates the production of thymus and activation regulated chemokine (TARC) and macrophage-derived chemokine (MDC), thus promoting the recruitment of Th2 effector cells into the lungs (Kohl et al., 2006). Data obtained from C5-deficient mice indicate that C5aR signaling is required to keep mDC susceptible to suppression T_{regs}, thereby controlling the immunogenic effect of mDC (Lewkowich et al., 2005).

A study by Zhang et al. demonstrates that regulation of co-stimulatory molecules on mDCs and pDCs is an important mechanism underlying the protective impact of C5aR signaling on the development of the Th2 immune response. HDM exposure increases the frequency of pulmonary mDCs expressing B7-H1 and B7-DC. In vivo ablation of C5aR signaling dramatically reduces the frequency of pDCs expressing B7-H1 and B7-DC. In the presence of C5aR signaling, blockade of B7-H1 or B7-DC results in enhanced Th2 cytokine production by T cells, suggesting that B7-H1 and B7-DC are critical for pDC-driven regulation of mDCs, and protection against allergic sensitization (Zhang et al., 2009). Interestingly, C5aR expression on dendritic cells might be downregulated in the Th2 cytokine-dominated environment, thus dampening the negative regulatory effect of C5aR signaling, in particular on pDCs. Indeed, high levels of IL-4 downregulates C5aR expression in monocyte-derived dendritic cells (Soruri et al., 2003).

In the absence of C5aR, C5a binds C5L2, which has a similar distribution in lung tissue as C5aR. The pro-allergic role of C5L2 in experimental asthma has been demonstrated recently. OVA-sensitized C5L2-deficient mice exhibit reduced AHR, inflammatory cell infiltration, and airway inflammation (Chen et al., 2007). Similarly, HDM-sensitized C5L2-deficient mice show significantly attenuated AHR and airway inflammation that is associated with decreased mucus production, Th2 cytokine production, and serum IgE levels (Zhang et al., 2010), confirming the pro-inflammatory role of C5L2 in the development of asthma. The available data further indicates that C5L2 also has a regulatory impact on mDCs. Adoptive transfer of C5L2-deficient mDCs into WT mice results in decreased eosinophilic and lymphocytic inflammation and a decreased IgE response. Importantly, C5L2-deficient mDCs still promote production of large amounts of Th2 cytokines from lung cells; however, in contrast to their WT counterparts, the production of Th1 and Th17 cytokines is also enhanced, and is associated with an increased neutrophil influx. Moreover, HDM

stimulation of C5L2-deficient mDCs results in production of the Th17-promoting cytokine IL-23. Collectively, this suggests a complex role of C5L2 in asthma; controlling the development of Th1 and Th17 cells in response to allergen challenge as well as driving the Th2 immune response (Zhang et al., 2010).

Th17 are IL-17A-producing subset of CD4⁺ T cells which have received much interest recently in allergy research. Several reports suggest that in addition to Th2 cells, Th17 cells contribute to the development of allergic inflammation. Sputum IL-17A mRNA is significantly elevated in asthmatic patients, and correlates with increase in sputum neutrophil counts (Bullens et al., 2006). Bronchial biopsies taken from patients with severe asthma show massive infiltration of IL-17 producing Th17 cells (Pene et al., 2008). In experimental asthma, IL-17A (Hellings et al., 2003) and Th17 (McKinley et al., 2008) cells have been shown to promote bronchial influx of neutrophils and induce airway inflammation and AHR. Th17 cells not only mediate neutrophilic airway inflammation, they also upregulate airway eosinophilia, together with IL-23 (Wakashin et al., 2008). Recent studies have highlighted an important dual role for C5a in Th17 cell development. C5aR-deficient mDCs show elevated production of Th17 promoting cytokines (transforming growth factor (TGF)- β , IL-6, and IL-23) and Th17 cell differentiation (Weaver et al., 2010). Furthermore, A/J mice (lacking C5) develop severe AHR associated with elevated levels of IL-17A and Th2 cytokines in comparison to C3H/HeJ strain of mice (mice manifesting less severe AHR). HDM challenge induces increased frequency of Th17 cells, as well as IL-17A in lungs of A/J mice. *In vivo* blockade of IL-17A with an anti-IL-17A antibody reduces AHR and BALF neutrophilia in HDM challenged A/J mice. Furthermore, an inverse relationship is found between the serum C5a concentration and the frequency of Th17 cells and IL-17A staining in lungs of HDM challenged A/J mice. Pharmacological inhibition of C5aR in BALB/c mice leads to enhanced influx of Th17 cells into the lungs after HDM challenge, and increased production of IL-17A per cell. Also, dendritic cells from these mice produce more HDM-driven IL-23. Taken together this suggests that C5a/C5aR complex controls IL-17A by limiting IL-23 production (Lajoie et al., 2010). As described above, Th17 development in experimental asthma is not only regulated by C5aR but by C5L2 as well (Zhang et al. 2010). In addition to its impact on Th17 cells, C5a may also regulate the development of T_{regs} (Palomares et al., 2010). Lack of C5aR signaling in mDCs results in T_{regs} differentiation (Lajoie et al., 2010). Thus, C5aR signaling in APCs can modulate Th cell differentiation at several levels in allergic asthma.

The development of asthma is not only regulated by C5a-C5aR/C5L2 interactions, but also by C3a-C3aR; however the role is less clear. In contrast to C5a, several studies suggest that C3a mainly contributes to the pathogenesis of asthma. When challenged with allergen, C3-deficient mice exhibit diminished AHR and lung eosinophilia (Drouin et al., 2001). C3aR-deficient mice challenged with *Aspergillus fumigatus* and OVA show attenuated/decreased AHR, airway eosinophilia, lung IL-4 producing cells, BALF levels of Th2 cytokines, serum IgE, and mucus production (Drouin et al., 2002). Similarly, C3a is also involved in the development of the late asthmatic response and AHR; the mechanism being related to the production of IL-1 β in the lung of OVA-challenged mice in response to C3a (Mizutani et al., 2009). However, in a model of particulate matter-induced pulmonary allergy, C3-deficient mice develop dramatically reduced AHR, but they are not protected from airway inflammation (Walters et al., 2002). Similarly, OVA-sensitized C3aR-deficient mice show markedly reduced bronchoconstriction and AHR, but IgE production and Th2 cytokine levels remains the same when compared to WT mice (Humbles et al., 2000).

Differences in mice strains, nature of the allergens used for sensitization and route of allergen administration may account for the conflicting results generated in different animal models. The study by Zhang et al. compared Th2 immune responses in C5aR- and C3aR-deficient mice side-by-side. As mentioned earlier, in the absence of C5aR signaling HDM-challenged mice suffer from an enhanced Th2 immune response. However, C3aR-deficient mice are protected against a Th2 immune response under same settings, delineating the opposing roles of C5aR and C3aR signaling in asthma. Interestingly, blocking C5aR-mediated signaling in C3aR-deficient mice significantly increases AHR, airway eosinophilia, and Th2 cytokines production, indicating that the decreased Th2 immune response in the absence of C3aR signaling results from a shift towards protective C5aR signaling (Zhang et al., 2009). Cross-talk between the two receptors is further supported by the fact that C5a negatively regulates C3aR internalization. Pulmonary C5aR-deficient dendritic cells exhibit dramatically higher expression of C3aR in comparison to WT dendritic cells. In contrast, C5aR expression significantly decreases in HDM-pulsed WT dendritic cells, but less so in C3aR-deficient dendritic cells, further confirming that there is a reciprocal regulation between C5aR and C3aR (Settmacher et al., 1999).

Recently, it has been shown that AHR, lung eosinophilia and mucus production are significantly increased in C5-deficient mouse model of Respiratory Syncytial Virus disease. C3aR expression in bronchial epithelial and smooth muscle cells of these mice are elevated as compared with WT mice. Ablation of C3aR signaling in C5-deficient mice significantly attenuates the disease phenotype, suggesting that C5 plays a crucial role in modulating AHR and eosinophilic inflammation by affecting expression of C3aR in the lungs (Melendi et al., 2007). C3a also has an impact on Th17 cell development. C3aR-deficient mice exhibit diminished AHR, fewer Th17 cells and decreased IL-17 compared to WT counterparts, which correlates with reduced IL-23 production. This suggests that unlike C5a, C3a mediates IL-23 production, IL-17 production and susceptibility to AHR (Lajoie et al., 2010).

It is clear that in experimental models of allergic asthma, C5a-C5aR signaling seems to protect against the development of Th2 immune response during allergen exposure, whereas C3a-C3aR signaling contributes to the development of maladaptive immune responses. However, the strong evidence that suggests C5a and C3a synergistically contribute to the development of allergic inflammation and asthma can not be overlooked. As mentioned earlier, the contradictory nature of this evidence may be due to the fact that once allergic inflammation is established (effector phase of allergic asthma), both C3a and C5a act on circulating and tissue resident inflammatory immune cells such as mast cells, eosinophils, basophils, and lymphocytes leading to the induction of a pro-allergic immune response. Indeed, pharmacological targeting of C3aR and C5aR during the effector phase suppresses AHR, airway inflammation and Th2 cytokines production (Baelder et al., 2005). Thus, complement C3a and C5a, and their receptors display diverse activities during the course of disease progression. Reagents that specifically targets C3a, C3aR, C5, C5a or C5aR could serve as potential therapy for asthma.

5.2 Sepsis

Sepsis is a life-threatening medical condition characterized by dysregulated systemic inflammatory responses followed by immunosuppression. Despite advances in medical health care, sepsis remains one of the leading causes of death, accounting for more than 1.5 million deaths annually in Europe and North America. Systemic inflammation in sepsis can

be triggered by various infectious agents, including bacteria (leading cause of sepsis), fungi, parasites and viruses. Over recent years, efforts to better understand the pathophysiology of sepsis, has given rise to enough convincing evidence to suggest that the activation of the complement system and production of C3a and C5a occurs in sepsis.

Infectious agents activate the complement system through their interaction with C1q, MBL, or ficolins leading to local and/or systemic complement activation with subsequent production of C3a and C5a. Indeed, patients with sepsis syndrome show elevated plasma or serum levels of C3a/C3a desArg, C4a and C5a/C5a desArg (Bengtson and Heideman, 1988, Cole et al., 2002, Nakae et al., 1994, Nakae et al., 1996, Selberg et al., 2000, Solomkin et al., 1985, Weinberg et al., 1984), and circulating levels of C3a and C5a are inversely proportional to patient survival (Groeneveld et al., 2003, Hack et al., 1989, Hecke et al., 1997, Selberg et al., 2000). In humans, C5a levels can rise as high as 100 nM (Ward, 2004).

In vivo generation of C3a and C5a and their inflammatory effects in sepsis have been studied using three major animal models: a) intravenous injection of an exogenous toxin (e.g. anaphylatoxins); b) intravenous infusion of exogenous bacteria; and c) cecal ligation and puncture (CLP). Infusion of C5a into rabbits and rats produces the typical septic shock symptoms, including a rapid drop in mean arterial pressure and reduced circulation of granulocytes, monocytes and platelets in peripheral blood. Infusion of *Escherichia coli* (*E. coli*) in rabbits leads to severe septicemia with enhanced levels of C5a in the plasma, indicating that the degree of complement activation determines the severity of sepsis (Bergh et al., 1991). Administration of an anti-C5a desArg antibody to *E. coli*-infused primates confers protection against mortality and attenuates systemic manifestations of sepsis (Stevens et al., 1986), highlighting the importance of C5a in the septic shock pathology. Most of the studies dealing with the role of C5a in pathology of sepsis have been done using the rat and rodent models of CLP. This model of sepsis closely mimics the pathophysiology of sepsis in humans. Intravenous infusion of a neutralizing polyclonal antibody to rat C5a in CLP rats is highly protective, causing reduced evidence of multiorgan failure and resulting improved survival rates (from 0% survival in the unprotected to 50% survival in rats treated with an anti-C5a antibody). Moreover, CLP rats treated with an antibody against C5a show profound reduction (98% compared to unprotected rats) in bacterial colony forming units (CFUs), indicating that C5a is associated with the development of bacteremia in sepsis (Czermak et al., 1999).

Successive studies with anti-C5a antibody-treated CLP rats and mice have further revealed that the survival rate of these animals depends on amount of antibody infused at the time of CLP, as well as the time of administration of the antibody. For instance, the infusion of 600 g of antibody targeted against the mid region of C5a remarkably improves survival in CLP rats (83-90% survival rate compared to 23% survival rate in CLP rats receiving pre-immune IgG or 100-220 g of anti-C5a antibody). Moreover, when 600 g of antibody is given at time 0 of CLP, the survival rate is 90%; when infusion is delayed until 6 hr. post CLP, survival is around 60%; and when delayed until 12 hr. post CLP, the survival drops to 40% (Huber-Lang et al., 2001). These results suggest that neutralization of C5a during a specific time window after the onset of sepsis may be efficacious in the treatment of sepsis.

In sepsis, excessive production of C3a and C5a subsequently leads to dysfunction of neutrophils. Blood neutrophils display suppressed chemotactic responsiveness (Goya et al., 1994, Solomkin et al., 1981), depressed enzyme release (Goya et al., 1994, Solomkin et al., 1981, Utoh et al., 1989), alteration of intracellular pH (Sachse et al., 2000), and a defective respiratory burst (Czermak et al., 1999, Goya et al., 1994, Solomkin et al., 1981), resulting in

impaired bactericidal activity. For instance, during experimental sepsis, blood neutrophils show a decreased ability to bind C5a, impaired chemotactic response to C5a and a loss of H₂O₂-generating capacity. Exposure of rat neutrophils to C5a induces a defect in phagocytic function (Huber-Lang et al., 2002). Treatment of CLP rats with an anti-C5a antibody reverses these functional defects in neutrophils (Huber-Lang et al., 2002). Early after CLP, blood neutrophils from septic mice exhibit enhanced expression of the CC chemokine receptors (CCR1, CCR2, and CCR5) (Speyer et al., 2004), which may contribute to a stronger neutrophilic inflammatory response in sepsis. Moreover, CLP causes blood neutrophils to show an increased expression of β 1 integrins (CD29) and β 2 integrins (CD18), indicating a hyperresponsiveness to the receptors for these integrins (Guo et al., 2002). Collectively, this demonstrates that neutrophils develop an exaggerated response to various inflammatory mediators in the early stages of sepsis.

Besides neutrophil dysfunction, C5a also affects other components of innate immunity leading to exacerbation of septicemia and immunosuppression. Systemic activation of complement in CLP model of sepsis induces C5a-dependent apoptosis of thymocytes (Guo et al., 2000), inducing a significant loss of thymus in first few days following CLP (Riedemann et al., 2002). C5aR expression on thymocytes rises rapidly after CLP and increased binding of C5a to CLP thymocytes can be found as early as 3 hr. post CLP. Administration of an anti-C5a neutralizing antibody at the time of CLP preserves the thymic mass and abolishes the apoptosis of thymocytes (Riedemann et al., 2002). Moreover, CLP in mice induces significant apoptosis of adrenomedullary cells 24 hr. post CLP, which is diminished after dual blockade of both C5aR and C5L2 (Flierl et al., 2008), indicating a role of C5a in multiorgan apoptosis during sepsis. C5a also promotes septic cardiomyopathy (Niederbichler et al., 2006) and consumptive coagulopathy (Laudes et al., 2002). The intensity of the coagulopathy of sepsis is greatly attenuated in CLP rats (for example, clotting times are minimally prolonged, thrombocytopenia is reduced and plasma levels of fibrin split products as well as thrombin-antithrombin complexes are greatly reduced) as a result of neutralization of C5a after CLP (Laudes et al., 2002).

It seems clear that excessive C5a produced during sepsis has harmful effects, as described above and it is obvious that the effects are mediated via the interaction of C5a with its receptors. Indeed, experimental studies in animals with CLP suggest a dynamic balance between C5aR and C5L2 on immune cells and in organs. C5aR expression is markedly increased in lung, liver, kidney, and heart early in septic mice (Riedemann et al., 2002). C5L2 mRNA is highly expressed in liver and thyroid of CLP rat, and weakly expressed in CLP rat brain, spleen, kidney, large intestine, eye as well as lung alveolar macrophages and peripheral blood neutrophils (Gao et al., 2005). Increased expression of C5aR and C5L2 in lung, liver, heart, and kidney after CLP-induced sepsis strongly implies the role of complement receptors in multiple organ failure.

In vitro exposure of neutrophils to C5a reduces surface C5aR expression suggesting that following interaction C5a/C5aR complex undergo internalization, suggesting a possible cause for compromised neutrophil function (Huber-Lang et al., 2002). During experimental sepsis C5aR content on blood neutrophils reduces 2.5-fold 24 hr. after CLP and increases steadily thereafter. On contrary, C5L2 content on blood neutrophils increases significantly 24 and 36 hr. after onset of CLP. Moreover, confocal microscopy on blood neutrophils from CLP rats show cytoplasmic distribution of C5aR, but not C5L2, indicating that C5a/C5L2 complex do not undergo internalization as C5a/C5aR after CLP (Gao et al., 2005), and that C5aR and C5L2 are independently regulated during sepsis. In presence of a cyclic peptide

antagonist (C5aRa) to the C5aR, the binding of C5a to mice peritoneal neutrophils is diminished, and the *in vitro* chemotactic response of neutrophils to C5a is decreased, C5a-induced defect in the oxidative burst of neutrophils is reversed, and the lung vascular permeability index is markedly diminished. CLP mice treated with C5aRa show improved survival rates (from 10% survival in the sham to 60% survival in mice treated with C5aRa) (Huber-Lang et al., 2002). Using a combination of C5aR and C5L2 blocking antibodies, and mice lacking one of the two C5a receptors, considerable improvement in survival following CLP has been shown in a study by Rittirsch et al. In mid-grade CLP, WT, C5aR-deficient and C5L2-deficient mice show 31%, 80% and 100% survival by day 7, respectively, indicating harmful roles for both C5a receptors in the CLP model of sepsis. All WT mice show 100% and 80% survival after treatment with anti-C5aR and anti-C5L2 serum, respectively, compared to 40% survival in mice treated with preimmune serum. In high-grade CLP, none of the WT mice treated with preimmune serum, C5L2-deficient mice treated with anti-C5aR serum and C5aR-deficient mice survived. Interestingly, when C5L2-deficient mice are treated with anti-C5aR serum the survival rate improves significantly (80%). Similarly, treatment of C5aR-deficient mice with anti-C5L2 serum greatly improves survival (87% compared to 17% for C5aR-deficient mice treated with preimmune serum or 0% for WT mice treated with anti-C5L2 serum). Dual blockade of C5aR and C5L2 by a polyclonal antibody is also protective against the lethal outcome after high-grade CLP. Moreover, if the high-grade CLP mice are treated with anti-C5aR and anti-C5L2 serum after a delay of 12 hr. or 24 hr. post CLP there is no protective effect against mortality. The combined blockade of the C5a receptors during sepsis is most effective when given before the onset of sepsis. In summary, both C5aR and C5L2 contribute cooperatively to mortality in sepsis. Evaluation of plasma cytokine and chemokine concentrations after CLP shows a reduction in the levels of IL-1 β , MIP-1 α , and MIP-2 in C5aR-deficient and C5L2-deficient mice in comparison to WT mice. However, the plasma concentration of IL-6 is significantly increased in C5L2-deficient CLP mice in comparison to WT mice. In contrast, plasma IL-6 is markedly reduced in C5aR-deficient CLP mice when compared to WT mice, as observed by Gao et al. (Gao et al., 2005), suggesting that the sepsis-induced cytokine and chemokine production may occur in a sequential fashion which requires C5a engagement of both C5a receptors. Another important observation obtained from this study is that C5L2-deficient CLP mice have greatly reduced levels of high-mobility group protein 1 (HMGB1, a late mediator in sepsis) in plasma. Interestingly, CLP in C5aR-deficient CLP mice has no effect on plasma HMGB1 levels when compared to WT CLP mice. Furthermore, C5a-stimulated macrophages from WT and C5aR-deficient mice produce significant amounts of HMGB1, whereas C5a-stimulated macrophages from C5L2-deficient mice produce very little HMGB1 (Rittirsch et al., 2008), supporting the fact that C5a mediates the pathophysiology of sepsis by acting on C5a receptors and that C5L2 has a functional role in sepsis.

Recently, a study has shown that $\gamma\delta$ T cells are involved in the pathogenesis of sepsis by producing large amounts of IL-17 (Xu et al., 2010), express C5aR, and the expression of C5aR increases in mice following sepsis. Neutralization of C5a partially prevents the upregulation of C5aR on $\gamma\delta$ T cells in septic mice. Furthermore, C5a promotes the IL-17 expression by $\gamma\delta$ T cells which can be attenuated by blocking PI3K/Akt signaling pathway, demonstrating that C5a acts on C5aR expressed on $\gamma\delta$ T cells, resulting in the pathophysiology of sepsis (Han et al.).

In summary, C5a binding to C5aR and C5L2 receptors seem to contribute to cytokine storm, associated multiple organ dysfunction and subsequent lethal outcome in the setting of experimental sepsis. C5aR and C5L2 both contribute synergistically to the harmful events in

sepsis. A maximal beneficial effect can be achieved by the blockade or absence of both receptors, which might have implication in complement-based therapy for inflammatory diseases.

5.3 Chronic urticaria

Chronic urticaria is a debilitating skin condition characterized by the near daily occurrence of pruritic wheals for at least six weeks (Kaplan, 2004). While the pathogenesis of chronic urticaria is not completely understood, mast cell and basophils degranulation and histamine release are believed to be of central importance. Recent studies suggest that this activation of mast cells and basophils could in part be initiated by the C3a and C5a or these complement proteins can augment allergen-antibody mediated cell activation. Indeed, heating serum from patients with chronic urticaria, which heat-inactivates complement proteins, reduces the ability of serum to induce histamine release from basophils. Similarly, decomplexed sera deficient in C5 is incapable of releasing histamine from dermal mast cells (Kikuchi and Kaplan, 2002).

C5a may play a key role in the pathogenesis of chronic urticaria as it can degranulate mast cells and basophils following its interaction with the C5aR present on these cells (Fureder et al., 1995). C5a can also chemoattract neutrophils, basophils, eosinophils and mast cells, which are present in chronic urticaria lesions. Korosec et al. showed that patients with chronic urticaria have enhanced basophil activation in response to C5a. However, C5aR antagonist-treated serum from these patients show decreased histamine release from basophil. Furthermore, the release of histamine from basophils by anti-FcεRI autoantibodies can be augmented by C5a activation (Korosec et al., 2009). Taken together these studies suggest that complement proteins and their receptors contribute towards the pathology of chronic urticaria.

5.4 Cancer

The complement system has also been well recognized as a promoter of tumor development. The deposition of complement component C3 is associated with the tumor vasculature in mice; C3-deficient mice show reduced tumor growth. The anaphylatoxin C5a promotes the growth of malignant tumors in a mouse model of cervical carcinoma. C5aR-deficient mice treated with a C5a antagonist exhibit reduced tumor growth, as well as enhanced infiltration of tumor tissue with CD8⁺ cytotoxic T cells in comparison to WT mice, suggesting that C5a promotes tumor growth by inhibiting the response of CD8⁺ cytotoxic T cells against tumors. Elimination of CD8⁺ cytotoxic T cells from C5aR-deficient increases the rate of tumor growth, confirming that C5a modulates the CD8⁺ T cells-mediated anti-tumor immune responses (Markiewski et al., 2008).

The interplay between CD8⁺ cytotoxic T cells and myeloid-derived suppressor cells (MDSCs) play a significant role in determining the fate of tumors. MDSCs are a heterogeneous population of regular myeloid cells, also referred to as immature counterparts of monocytes and neutrophils, which express C5aR. C5aR is partially internalized in tumor-associated MDSCs, suggesting that MDSCs are constantly exposed to and activated by C5a generated in the tumor microenvironment. C5a chemoattracts MDSCs to the tumor, partly by upregulating adhesion molecules on MDSCs. C5a also enhances the production of ROS and reactive nitrogen species (RNS) from MDSC, which are known to suppress CD8⁺ cytotoxic T cell anti-tumor responses. C5aR-deficient mice or mice treated

with a C5aR antagonist show reduced numbers of MDSCs within the tumor tissue, increased numbers of CD8⁺ cytotoxic T cells and decreased tumor growth. MDSCs from C5a-deficient mice are unable to inhibit T cell proliferation *in vivo* (Markiewski et al., 2008). These observations highlight the potential of anaphylatoxins and their receptors as novel targets for anti-cancer immunotherapy.

5.5 Ischemia-reperfusion injury

Ischemia-reperfusion (I/R) injury is defined as cellular injury occurring after the reperfusion of previously vascularised tissue following an extended period of ischemia. The augmentation of tissue injury after reperfusion results from an intense inflammatory response that develops simultaneously with tissue reperfusion (Eltzschig and Collard, 2004). Several pathological conditions can lead to I/R injury including myocardial infarction, stroke, hemorrhagic shock, severe trauma, and organ transplantation resulting in associated morbidity and mortality (Eltzschig and Collard, 2004).

Numerous studies have shown that ischemic tissue activates the complement system, which remarkably contributes to the development of tissue damage by enhancing inflammation (Hart et al., 2004). The first evidence for involvement of complement in I/R injury was proposed by Hill and Ward in 1971 (Hill and Ward, 1971). During I/R injury the complement system can be activated by the classical, alternative, and lectin pathways. For instance, skeletal muscle injury resulting from I/R likely occurs through the complement activation via the classical and lectin pathways (Weiser et al., 1996). Lectin pathway may be involved in myocardial (Jordan et al., 2001) and gastrointestinal I/R-induced complement activation (Hart et al., 2004). However, the amplification of complement activation in gastrointestinal I/R occurs through the alternative pathway (Hart et al., 2004). Alternative pathway of complement activation may contribute to renal I/R injury in mice (Thurman et al., 2003).

Studies suggest that complements C3a and C5a are major complement factors responsible for the induction of the reperfusion-associated inflammatory response. C3a and C5a activate endothelial cells and inflammatory leukocytes. C5a upregulates the expression of adhesion molecules on human umbilical vein endothelial cells (Foreman et al., 1996) and induces release of various cytokines, including IL-1, IL-6, MCP-1, and TNF α (Schindler et al., 1990). C5aR expression is upregulated following cold I/R injury in a mouse model of syngenic kidney transplantation, suggesting that C5aR may contribute to tissue damage, tubular apoptosis and dysfunction of donor organs. Furthermore, upregulation of C5aR expression in cadaveric kidneys correlates with cold ischemia time. Ablation of C5aR signaling during cold ischemia has a protective effect on kidney graft survival (Lewis et al., 2008). Animals treated with a C5aR antagonist show dramatically reduced accumulation of neutrophils in the post-ischemic livers and sustain less injury during reperfusion (Arumugam et al., 2003). In a mouse model of I/R muscle injury, mice treated with a C5aR antagonist exhibit decreased levels of circulating creatinine kinase, lactate dehydrogenase, alanine and aspartate aminotransferase, creatinine, blood urea nitrogen, muscle edema, lung and liver myeloperoxidase, and lung TNF α following hind limb injury (Woodruff et al., 2004).

Studies targeting C5a/C5aR complex have further confirmed the role played by C5a in the pathogenesis of I/R injury. Blocking C5aR signaling using an anti-C5aR antibody markedly decreases leukocyte adherence, microvascular permeability in the ischemic myocardial area (Zhang et al., 2007), myocardial neutrophil infiltration and arteriolar endothelial injury

(Park et al., 1999). Knocking down C5aR expression with small interference (si)RNA preserves renal function from I/R injury, reduces chemokine production and neutrophil infiltration (Zheng et al., 2008). Treatment with an anti-C5 neutralizing antibody reduces apoptosis and necrosis in heart allografts (Ferraresso et al., 2008).

In contrast to C5a, the role of C3a/C3aR in I/R injury is not properly established. Systemic inhibition of C3a with a C3aR antagonist minimally resolves myocardial I/R injury, and neutropenia rather than C3aR antagonism appears to be responsible for C3aR antagonist-associated improvement in myocardial I/R injury (Busche and Stahl, 2010). These results confirm similar observations from previous studies (Ames et al., 2001, Proctor et al., 2004), indicating that C3aR antagonist-mediated neutrophil tissue sequestration during I/R injury may account for the protective effects observed. Overall, the data indicate while C3a/C3aR inhibition in the clinical setting of I/R injury does not appear to be therapeutic, targeting C5a as well as C5aR may be a promising approach to prevent I/R injury.

5.6 Transplantation

The activation products of complement system play an important role in allograft rejection as evidenced by the fact that the lack of C3 in donor kidneys is associated with long-term graft survival in experimental transplantation (Pratt et al., 2002). A study in human kidney transplantation has shown that donor C3 polymorphisms are associated with late graft failure. Thus donor expression of C3 influences the alloimmune response and the fate of the transplantation (Brown et al., 2006). APCs are the source of C3 and macrophages from C3-deficient mice have an impaired potency to stimulate alloreactive T cell response and to drive Th1-biased adaptive immune responses supporting graft survival (Peng et al., 2006, Zhou et al., 2006). It is now clear that this effect of C3 is mediated via the interaction of C3a with the C3aR on APCs. C3a-deficient dendritic cells show reduced surface expression of major histocompatibility complex (MHC) class II and co-stimulatory molecules and elicit a defective T cell priming against alloantigen expressed on the dendritic cells (Peng et al., 2008). Mechanistically, C3a-C3aR interaction on surface of dendritic cells decreases the level of intracellular cAMP, which in turn promotes allergen uptake and T cell priming (Li et al., 2008).

As mentioned in previous section, complement activation is critically involved in I/R injury; C5a and C5aR blockade has been shown attenuate organ damage, improve graft function and transplantation outcome. In human kidney transplants with acute rejection, C5aR expression is increased in renal tissue and in cells infiltrating the tubulointerstitium. Treatment of recipient mice with a C5aR antagonist before transplantation markedly improves renal allograft survival and reduces alloreactive T cell priming. Similarly, inhibition of C5aR in murine model of renal allotransplantation substantially improved graft survival from 11 days to 12 weeks. In addition, C5aR inhibition reduces kidney inflammation, apoptosis, and priming of alloreactive T cells (Gueler et al., 2008). Pharmacological targeting of C5aR during organ preservation significantly improves kidney graft survival (Lewis et al., 2008). Baboons treated with an anti-C5a monoclonal antibody exhibit prolonged pulmonary xenografts survival, indicating that C5a exacerbates pulmonary xenografts injury (Gaca et al., 2006). A recent study by Vieyra et al. have shown that dendritic cell-derived C3a and C5a regulates CD4⁺ T cell help to CD8⁺ T cell responses required for murine allograft rejection (Vieyra et al., 2011). These results demonstrate that C3aR and C5aR signaling contribute towards the innate and adaptive inflammatory

responses following solid organ transplantation, suggesting that pharmaceutical targeting of C3aR and C5aR may have an application in transplantation medicine.

5.7 Stroke

Stroke is the second leading cause of death worldwide, which is defined as a rapidly developing loss of brain function(s) due to inadequate blood flow in a localized area. When blood flow is interrupted to part of the brain, brain cells quickly begin to die leading to stroke development. There are many elements that contribute to the development of stroke, of which neuroinflammation is a major one. However, the precise roles of various pro-inflammatory mediators, including cytokines, chemokines and immune cells, are still largely unknown. Increasing evidence suggests that the activation of the complement cascade contributes to pathological inflammatory events in brain (upregulation of adhesion molecules, immune cell activation, chemotaxis, expression of IL-8 and MCP-1 by endothelial cells) (D'Ambrosio et al., 2001), resulting in ischemic insult, neurodegeneration and stroke development.

Following brain I/R injury ROS, RNS as well as oxygen free radicals are generated by activated brain cells and infiltrating immune cells, which stimulate stroke-associated brain injury (Elsner et al., 1994, McColl et al., 2009). Brain cells such as astrocytes, microglia, neurons, and endothelial cells and infiltrating immune cells produce various pro-inflammatory mediators following ischemia, further contributing towards cell death (Yilmaz et al., 2006). Ischemic stroke enhances interaction between endothelial cells, brain cells, and immune cells that may aggravate the injury process (Urrea et al., 2009). All these stroke-associated brain pathologies are function of C3a-C3aR and C5a-C5aR interactions (D'Ambrosio et al., 2001). Strong C3a and C5a activation is observed in patients with acute ischemic stroke, which correlates with disease severity (Szeplaki et al., 2009). Regional brain I/R injury induces an inflammatory reaction that involves generation of C3a and C5a, upregulation and enhanced activation of their receptors C3aR and C5aR.

In the brain, C3aR and C5aR are expressed by astrocytes (Gasque et al., 1995, Gasque et al., 1997, Gasque et al., 1998, Lacy et al., 1995, Sayah et al., 1997), glial cells (Davoust et al., 1999, Gasque et al., 1997, Lacy et al., 1995) and neurons (Davoust et al., 1999). Experimental models of permanent and transient middle cerebral artery occlusion (MCAO) have demonstrated an increase in the expression of complement receptors. The expression of C3aR and C5aR is enhanced in mouse ischemic cortex following permanent MCAO. In addition, the expression is also induced on endothelial cells within ischemic core, suggesting that the complement receptors are important in leukocyte recruitment and neuroinflammation (Van Beek et al., 2000). Expression of C3aR and C5aR is significantly increased after transient MCAO in rats (Nishino et al., 1994) and mice (Barnum et al., 2002). In the later study, C5aR expression dramatically increases within 3 hr. after MCAO, whereas C3aR expression reduces to 25% of control animals. By 24 hr. post-occlusion, expression of both receptors is highest. This increased expression at the later time points after occlusion is most likely the result of a massive infiltration of immune cells expressing C3aR and C5aR (Barnum et al., 2002).

Early attempts at complement inhibition using cobra venom factor (CVF) have further revealed that abrogation of the complement system can provide protection for the brain during I/R injury and stroke. Pretreatment of rats with CVF (complement-depleted rats) prior to temporary cerebral ischemia significantly enhances the magnitude of reactive

hyperemia and increases preservation of somatosensory evoked potentials (SSEPs), demonstrating that depleting the complement system can improve blood flow and clinical outcome following cerebral I/R (Vasthare et al., 1998). Similarly, complement depletion via CVF significantly reduces post-ischemic cerebral infarct volume in adult rats and post-hypoxic-ischemic cerebral atrophy in neonates (Figuerola et al., 2005). However, the lack of specificity of CVF did not give any information regarding complement components that are most relevant in the pathogenesis of brain injury and stroke.

Recent studies using genetic knockouts of C3 and C5 and inhibitors of C3a-C3aR, and C5a-C5aR signaling have better our understanding of the role of anaphylatoxins in stroke. C3-deficient mice are protected against cerebral I/R injury, as demonstrated by significant reductions in both infarct volume and neurological deficit score. C3-deficient mice also exhibit diminished granulocyte infiltration and oxidative stress. The administration of a C3aR antagonist reduces stroke volume leading to neurological improvement (Mocco et al., 2006), implicating the involvement of C3a and C3aR in acute stroke. Studies with genetic knockouts of C5 in stroke have yielded conflicting data. C5-deficient animals show increased vulnerability to intracerebral hemorrhage (ICH) (Nakamura et al., 2004) and ischemic stroke (Mocco et al., 2006). In contrast, C5-deficient mice subjected to brain I/R injury exhibits improved functional outcome and less brain damage (Arumugam et al., 2007). A recent study by Rynkowski et al. have shown that blocking C3a-mediated signaling using a C3aR antagonist is protective in a mouse model of ICH (Rynkowski et al., 2009). Similarly, mice treated with C5aR antagonist alone or C3aR and C5aR antagonists exhibit improvement in neurological functions following ICH, suggesting that blockade of C3aR and C5aR represents a promising therapeutic strategy in stroke.

6. Conclusion

The complement system is composed of a network of proteins that play an important role in innate and adaptive immunity. Originally discovered as antimicrobial agents, the main function of C3a and C5a was considered to be the opsonization of pathogens and chemoattraction to remove apoptotic and necrotic cells. However, today complement proteins, C3a and C5a, are considered as crucial immunoregulatory molecules with pleiotropic biological functions on immune cells which help to shape the immune response. Activation of complement system is exquisitely regulated, while improper activation or under certain conditions the effect can lead to adverse consequences. Similar to dysregulation of the adaptive immune system in hypersensitivity reactions, the pathological role of C3a, C5a and their receptors in inflammatory diseases as well as tumor growth is well defined. Due to their strong inflammatory properties, C3a/C3aR and C5a/C5aR are considered attractive pharmacological targets for the development of therapeutic agents. Given that many therapeutic agents targeting the interaction of C3a-C3aR and C5a-C5aR are already under investigation, the advances made in the field of complement and complement receptors discussed in this book chapter will better our understanding of the disease process and help develop new therapeutic approaches to modulate immune response

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8. References

- A genome-wide search for asthma susceptibility loci in ethnically diverse populations. The Collaborative Study on the Genetics of Asthma (CSGA). *Nature Genetics*, Vol.15, No.4, (April 1997), pp. 389-392, ISSN 1061-4036
- Abe, M., Shibata, K., Akatsu, H., Shimizu, N., Sakata, N., Katsuragi, T. and Okada, H. (2001). Contribution of anaphylatoxin C5a to late airway responses after repeated exposure of antigen to allergic rats. *Journal of Immunology*, Vol.167, No.8, (October 2001), pp. 4651-4660, ISSN 0022-1767
- Ahamed, J., Haribabu, B. and Ali, H. (2001). Cutting edge: Differential regulation of chemoattractant receptor-induced degranulation and chemokine production by receptor phosphorylation. *Journal of Immunology*, Vol.167, No.7, (October 2001), pp. 3559-3563, ISSN 0022-1767
- Albrecht, E. A., Chinnaiyan, A. M., Varambally, S., Kumar-Sinha, C., Barrette, T. R., Sarma, J. V. and Ward, P. A. (2004). C5a-induced gene expression in human umbilical vein endothelial cells. *The American Journal of Pathology*, Vol.164, No.3, (March 2004), pp. 849-859, ISSN 0002-9440
- Amatruda, T. T., 3rd, Gerard, N. P., Gerard, C. and Simon, M. I. (1993). Specific interactions of chemoattractant factor receptors with G-proteins. *Journal of Biological Chemistry*, Vol.268, No.14, (May 1993), pp. 10139-10144, ISSN 0021-9258
- Ames, R. S., Li, Y., Sarau, H. M., Nuthulaganti, P., Foley, J. J., Ellis, C., Zeng, Z., Su, K., Jurewicz, A. J., Hertzberg, R. P., Bergsma, D. J. and Kumar, C. (1996). Molecular cloning and characterization of the human anaphylatoxin C3a receptor. *Journal of Biological Chemistry*, Vol.271, No.34, (August 1996), pp. 20231-20234, ISSN 0021-9258
- Ames, R. S., Lee, D., Foley, J. J., Jurewicz, A. J., Tornetta, M. A., Bautsch, W., Settmacher, B., Klos, A., Erhard, K. F., Cousins, R. D., Sulpizio, A. C., Hieble, J. P., McCafferty, G., Ward, K. W., Adams, J. L., Bondinell, W. E., Underwood, D. C., Osborn, R. R., Badger, A. M. and Sarau, H. M. (2001). Identification of a selective nonpeptide antagonist of the anaphylatoxin C3a receptor that demonstrates antiinflammatory activity in animal models. *Journal of Immunology*, Vol.166, No.10, (May 2001), pp. 6341-6348, ISSN 0022-1767
- Arroyave, C. M., Stevenson, D. D., Vaughan, J. H. and Tan, E. M. (1977). Plasma complement changes during bronchospasm provoked in asthmatic patients. *Clinical Allergy*, Vol.7, No.2, (March 1977), pp. 173-182, ISBN 978-1-58829-616-0
- Arumugam, T. V., Shiels, I. A., Strachan, A. J., Abbenante, G., Fairlie, D. P. and Taylor, S. M. (2003). A small molecule C5a receptor antagonist protects kidneys from ischemia/reperfusion injury in rats. *Kidney International*, Vol.63, No.1, (January 2003), pp. 134-142, ISSN 0085-2538
- Arumugam, T. V., Shiels, I. A., Woodruff, T. M., Granger, D. N. and Taylor, S. M. (2004). The role of the complement system in ischemia-reperfusion injury. *Shock*, Vol.21, No.5, (May 2004), pp. 401-409, ISSN 1073-2322
- Arumugam, T. V., Tang, S. C., Lathia, J. D., Cheng, A., Mughal, M. R., Chigurupati, S., Magnus, T., Chan, S. L., Jo, D. G., Ouyang, X., Fairlie, D. P., Granger, D. N.,

- Vortmeyer, A., Basta, M. and Mattson, M. P. (2007). Intravenous immunoglobulin (IVIG) protects the brain against experimental stroke by preventing complement-mediated neuronal cell death. *Proceedings of the National Academy of Science of the United States of America*, Vol.104, No.35, (August 2007), pp. 14104-14109, ISSN 0027-8424
- Baelder, R., Fuchs, B., Bautsch, W., Zwirner, J., Kohl, J., Hoymann, H. G., Glaab, T., Erpenbeck, V., Krug, N. and Braun, A. (2005). Pharmacological targeting of anaphylatoxin receptors during the effector phase of allergic asthma suppresses airway hyperresponsiveness and airway inflammation. *Journal of Immunology*, Vol.174, No.2, (January 2005), pp. 783-789, ISSN 0022-1767
- Baldo, A., Sniderman, A. D., St-Luce, S., Avramoglu, R. K., Maslowska, M., Hoang, B., Monge, J. C., Bell, A., Mulay, S. and Cianflone, K. (1993). The adipsin-acylation stimulating protein system and regulation of intracellular triglyceride synthesis. *The Journal of Clinical Investigation*, Vol.92, No.3, (September 1993), pp. 1543-1547, ISSN 0021-9738
- Bamberg, C. E., Mackay, C. R., Lee, H., Zahra, D., Jackson, J., Lim, Y. S., Whitfeld, P. L., Craig, S., Corsini, E., Lu, B., Gerard, C. and Gerard, N. P. (2010). The C5a receptor (C5aR) C5L2 is a modulator of C5aR-mediated signal transduction. *Journal of Biological Chemistry*, Vol.285, No.10, (March 2010), pp. 7633-7644, ISSN 0021-9258
- Banchereau, J. and Steinman, R. M. (1998). Dendritic cells and the control of immunity. *Nature*, Vol.392, No.6673, (March 1998), pp. 245-252, ISSN 0028-0836
- Bansal, G., Xie, Z., Rao, S., Nocka, K. H. and Druey, K. M. (2008). Suppression of immunoglobulin E-mediated allergic responses by regulator of G protein signaling 13. *Nature Immunology*, Vol.9, No.1, (January 2008), pp. 73-80, ISSN 1529-2908
- Barnum, S. R., Ames, R. S., Maycox, P. R., Hadingham, S. J., Meakin, J., Harrison, D. and Parsons, A. A. (2002). Expression of the complement C3a and C5a receptors after permanent focal ischemia: An alternative interpretation. *Glia*, Vol.38, No.2, (April 2002), pp. 169-173, ISSN 1098-1136
- Bautsch, W., Hoymann, H. G., Zhang, Q., Meier-Wiedenbach, I., Raschke, U., Ames, R. S., Sohns, B., Flemme, N., Meyer zu Vilsendorf, A., Grove, M., Klos, A. and Kohl, J. (2000). Cutting edge: guinea pigs with a natural C3a-receptor defect exhibit decreased bronchoconstriction in allergic airway disease: evidence for an involvement of the C3a anaphylatoxin in the pathogenesis of asthma. *Journal of Immunology*, Vol.165, No.10, (November 2000), pp. 5401-5405, ISSN 0022-1767
- Becker, S., Meuer, S., Hadding, U. and Bitter-Suermann, D. (1978). Platelet activation: a new biological activity of guinea-pig C3a anaphylatoxin. *Scandinavian Journal of Immunology*, Vol.7, No.2, pp. 173-180, ISSN 1365-3083
- Bengtson, A. and Heideman, M. (1988). Anaphylatoxin formation in sepsis. *Archives of Surgery (Chicago, Ill.: 1960)*, Vol.123, No.5, (May 1988), pp. 645-649, ISSN 0004-0010
- Bergh, K., Olsen, P. O., Halgunset, J. and Iversen, O. J. (1991). Complement activation and pulmonary dysfunction in experimental *Escherichia coli* septicemia. *Acta Anaesthesiologica Scandinavica*, Vol.35, No.3, (April 1991), pp. 267-274, ISSN 0001-5172
- Bischoff, S. C., de Weck, A. L. and Dahinden, C. A. (1990). Interleukin 3 and granulocyte/macrophage-colony-stimulating factor render human basophils responsive to low concentrations of complement component C3a. *Proceedings of the*

- National Academy of Science of the United States of America*, Vol.87, No.17, (September 1990), pp. 6813-6817, ISSN 0027-8424
- Bito, L. Z. (1977). Inflammatory effects of endotoxin-like contaminants in commonly used protein preparations. *Science*, Vol.196, No.4285, (April 1977), pp. 83-85, ISSN 0036-8075
- Bokisch, V. A. and Muller-Eberhard, H. J. (1970). Anaphylatoxin inactivator of human plasma: its isolation and characterization as a carboxypeptidase. *The Journal of Clinical Investigation*, Vol.49, No.12, (December 1970), pp. 2427-2436, ISSN 0021-9738
- Boulay, F., Mery, L., Tardif, M., Bouchon, L. and Vignais, P. (1991). Expression cloning of a receptor for C5a anaphylatoxin on differentiated HL-60 cells. *Biochemistry*, Vol.30, No.12, (March 1991), pp. 2993-2999, ISSN 0006-2960
- Bowser, C., Erstein, D. P., Silverberg, J. I., Nowakowski, M. and Joks, R. (2010). Correlation of plasma complement split product levels with allergic respiratory disease activity and relation to allergen immunotherapy. *Annals of Allergy, Asthma & Immunology*, Vol.104, No.1, (January 2010), pp. 42-49, ISSN 1081-1206
- Braun, L., Christophe, T. and Boulay, F. (2003). Phosphorylation of key serine residues is required for internalization of the complement 5a (C5a) anaphylatoxin receptor via a beta-arrestin, dynamin, and clathrin-dependent pathway. *Journal of Biological Chemistry*, Vol.278, No.6, (February 2003), pp. 4277-4285, ISSN 0021-9258
- Brown, K. M., Kondeatis, E., Vaughan, R. W., Kon, S. P., Farmer, C. K., Taylor, J. D., He, X., Johnston, A., Horsfield, C., Janssen, B. J., Gros, P., Zhou, W., Sacks, S. H. and Sheerin, N. S. (2006). Influence of donor C3 allotype on late renal-transplantation outcome. *The New England Journal of Medicine*, Vol.354, No.19, (May 2006), pp. 2014-2023, ISSN 0028-4793
- Bullens, D. M., Truyen, E., Coteur, L., Dilissen, E., Hellings, P. W., Dupont, L. J. and Ceuppens, J. L. (2006). IL-17 mRNA in sputum of asthmatic patients: linking T cell driven inflammation and granulocytic influx?. *Respiratory Research*, Vol.7, (November 2006), pp. 135, ISSN 1465-9921
- Burg, M., Martin, U., Bock, D., Rheinheimer, C., Kohl, J., Bautsch, W. and Klos, A. (1996). Differential regulation of the C3a and C5a receptors (CD88) by IFN-gamma and PMA in U937 cells and related myeloblastic cell lines. *Journal of Immunology*, Vol.157, No.12, (December 1996), pp. 5574-5581, ISSN 0022-1767
- Busche, M. N. and Stahl, G. L. (2010). Role of the complement components C5 and C3a in a mouse model of myocardial ischemia and reperfusion injury. *German Medical Science*, Vol.8, (September 2010), ISSN 1612-3174
- Cain, S. A. and Monk, P. N. (2002). The orphan receptor C5L2 has high affinity binding sites for complement fragments C5a and C5a des-Arg(74). *Journal of Biological Chemistry*, Vol.277, No.9, (March 2002), pp. 7165-7169, ISSN 0021-9258
- Castro, F. F., Schmitz-Schumann, M., Rother, U. and Kirschfink, M. (1991). Complement activation by house dust: reduced reactivity of serum complement in patients with bronchial asthma. *International Archives of Allergy and Applied Immunology*, Vol.96, No.4, pp. 305-310, ISSN 0002-5915
- Chao, T. H., Ember, J. A., Wang, M., Bayon, Y., Hugli, T. E. and Ye, R. D. (1999). Role of the second extracellular loop of human C3a receptor in agonist binding and receptor function. *Journal of Biological Chemistry*, Vol.274, No.14, (April 1999), pp. 9721-9728, ISSN 0021-9258

- Chen, N. J., Mirtsos, C., Suh, D., Lu, Y. C., Lin, W. J., McKerlie, C., Lee, T., Baribault, H., Tian, H. and Yeh, W. C. (2007). C5L2 is critical for the biological activities of the anaphylatoxins C5a and C3a. *Nature*, Vol.446, No.7132, (March 2007), pp. 203-207, ISSN 0028-0836
- Christophe, T., Rabiet, M. J., Tardif, M., Milcent, M. D. and Boulay, F. (2000). Human complement 5a (C5a) anaphylatoxin receptor (CD88) phosphorylation sites and their specific role in receptor phosphorylation and attenuation of G protein-mediated responses. Desensitization of C5a receptor controls superoxide production but not receptor sequestration in HL-60 cells. *Journal of Biological Chemistry*, Vol.275, No.3, (January 2000), pp. 1656-1664, ISSN 0021-9258
- Cianflone, K., Vu, H., Walsh, M., Baldo, A., and Sniderman, A. (1989). Metabolic response of Acylation Stimulating Protein to an oral fat load. *Journal of Lipid Research*, Vol.30, No.11, (November 1989), pp. 1727-1733, ISSN 0022-2275
- Cole, L., Bellomo, R., Hart, G., Journois, D., Davenport, P., Tipping, P. and Ronco, C. (2002). A phase II randomized, controlled trial of continuous hemofiltration in sepsis. *Critical Care Medicine*, Vol.30, No.1, (January 2002), pp. 100-106, ISSN 0090-3493
- Crass, T., Raffetseder, U., Martin, U., Grove, M., Klos, A., Kohl, J. and Bautsch, W. (1996). Expression cloning of the human C3a anaphylatoxin receptor (C3aR) from differentiated U-937 cells. *European Journal of Immunology*, Vol.26, No.8, (August 1996), pp. 1944-1950, ISSN 0014-2980
- Czermak, B. J., Sarma, V., Pierson, C. L., Warner, R. L., Huber-Lang, M., Bless, N. M., Schmal, H., Friedl, H. P. and Ward, P. A. (1999). Protective effects of C5a blockade in sepsis. *Nature Medicine*, Vol.5, No.7, (July 1999), pp. 788-792, ISSN 1078-8956
- D'Ambrosio, A. L., Pinsky, D. J. and Connolly, E. S. (2001). The role of the complement cascade in ischemia/reperfusion injury: implications for neuroprotection. *Molecular Medicine*, Vol.7, No.6, (June 2001), pp. 367-382, ISSN 1757-4684
- Daffern, P. J., Pfeifer, P. H., Ember, J. A. and Hugli, T. E. (1995). C3a is a chemotaxin for human eosinophils but not for neutrophils. I. C3a stimulation of neutrophils is secondary to eosinophil activation. *The Journal of Experimental Medicine*, Vol.181, No.6, (June 1995), pp. 2119-2127, ISSN 0022-1007
- Davoust, N., Jones, J., Stahel, P. F., Ames, R. S. and Barnum, S. R. (1999). Receptor for the C3a anaphylatoxin is expressed by neurons and glial cells. *Glia*, Vol.26, No.3, (May 1999), pp. 201-211, ISSN 1098-1136
- DeMartino, J. A., Van Riper, G., Siciliano, S. J., Molineaux, C. J., Konteatis, Z. D., Rosen, H. and Springer, M. S. (1994). The amino terminus of the human C5a receptor is required for high affinity C5a binding and for receptor activation by C5a but not C5a analogs. *Journal of Biological Chemistry*, Vol.269, No.20, (May 1994), pp. 14446-14450, ISSN 0021-9258
- DiScipio, R. G., Daffern, P. J., Jagels, M. A., Broide, D. H. and Sriramaraio, P. (1999). A comparison of C3a and C5a-mediated stable adhesion of rolling eosinophils in postcapillary venules and transendothelial migration in vitro and in vivo. *Journal of Immunology*, Vol.162, No.2, (January 1999), pp. 1127-1136, ISSN 0022-1767
- Drouin, S. M., Corry, D. B., Kildsgaard, J. and Wetsel, R. A. (2001). Cutting edge: the absence of C3 demonstrates a role for complement in Th2 effector functions in a murine model of pulmonary allergy. *Journal of Immunology*, Vol.167, No.8, (October 2001), pp. 4141-4145, ISSN 0022-1767

- Drouin, S. M., Corry, D. B., Hollman, T. J., Kildsgaard, J. and Wetsel, R. A. (2002). Absence of the complement anaphylatoxin C3a receptor suppresses Th2 effector functions in a murine model of pulmonary allergy. *Journal of Immunology*, Vol.169, No.10, (November 2002), pp. 5926-5933, ISSN 0022-1767
- Elsner, J., Oppermann, M., Czech, W. and Kapp, A. (1994). C3a activates the respiratory burst in human polymorphonuclear neutrophilic leukocytes via pertussis toxin-sensitive G-proteins. *Blood*, Vol.83, No.11, (June 1994), pp. 3324-3331, ISSN 0006-4971
- Eltzschig, H. K. and Collard, C. D. (2004). Vascular ischaemia and reperfusion injury. *British Medical Bulletin*, Vol.70, (October 2004), pp. 71-86, ISSN 0007-1420
- Elward, K., Griffiths, M., Mizuno, M., Harris, C. L., Neal, J. W., Morgan, B. P. and Gasque, P. (2005). CD46 plays a key role in tailoring innate immune recognition of apoptotic and necrotic cells. *Journal of Biological Chemistry*, Vol.280, No.43, (October 2005), pp. 36342-36354, ISSN 0021-9258
- Ember, J. A. and Hugli, T. E. (1997). Complement factors and their receptors. *Immunopharmacology*, Vol.38, No.1-2, (December 1997), pp. 3-15, ISSN 1567-5769
- Ferraresso, M., Macor, P., Valente, M., Della Barbera, M., D'Amelio, F., Borghi, O., Raschi, E., Durigutto, P., Meroni, P. and Tedesco, F. (2008). Posttransplant ischemia-reperfusion injury in transplanted heart is prevented by a minibody to the fifth component of complement. *Transplantation*, Vol.86, No.10, (November 2008), pp. 1445-1451, ISSN 0041-1337
- Figueroa, E., Gordon, L. E., Feldhoff, P. W. and Lassiter, H. A. (2005). The administration of cobra venom factor reduces post-ischemic cerebral injury in adult and neonatal rats. *Neuroscience Letters*, Vol.380, No.1-2, (May 2005), pp. 48-53, ISSN 0304-3940
- Flierl, M. A., Rittirsch, D., Chen, A. J., Nadeau, B. A., Day, D. E., Sarma, J. V., Huber-Lang, M. S. and Ward, P. A. (2008). The complement anaphylatoxin C5a induces apoptosis in adrenomedullary cells during experimental sepsis. *PLoS One*, Vol.3, No.7, (July 2008), pp. e2560, ISSN 1932-6203
- Foreman, K. E., Glovsky, M. M., Warner, R. L., Horvath, S. J. and Ward, P. A. (1996). Comparative effect of C3a and C5a on adhesion molecule expression on neutrophils and endothelial cells. *Inflammation*, Vol.20, No.1, (February 1996), pp. 1-9, ISSN 0360-3997
- Fureder, W., Agis, H., Willheim, M., Bankl, H. C., Maier, U., Kishi, K., Muller, M. R., Czerwenka, K., Radaszkiewicz, T., Butterfield, J. H., Klappacher, G. W., Sperr, W. R., Oppermann, M., Lechner, K. and Valent, P. (1995). Differential expression of complement receptors on human basophils and mast cells. Evidence for mast cell heterogeneity and CD88/C5aR expression on skin mast cells. *Journal of Immunology*, Vol.155, No.6, (September 1995), pp. 3152-3160, ISSN 0022-1767
- Gaca, J. G., Appel, J. Z., 3rd, Lukes, J. G., Gonzalez-Stawinski, G. V., Leshner, A., Palestrant, D., Logan, J. S., Love, S. D., Holzkecht, Z. E., Platt, J. L., Parker, W. and Davis, R. D. (2006). Effect of an anti-C5a monoclonal antibody indicates a prominent role for anaphylatoxin in pulmonary xenograft dysfunction. *Transplantation*, Vol.81, No.12, (June 2006), pp. 1686-1694, ISSN 0041-1337
- Gao, H., Neff, T. A., Guo, R. F., Speyer, C. L., Sarma, J. V., Tomlins, S., Man, Y., Riedemann, N. C., Hoesel, L. M., Younkin, E., Zetoune, F. S. and Ward, P. A. (2005). Evidence for a functional role of the second C5a receptor C5L2. *Faseb Journal*, Vol.19, No.8, (June 2005), pp. 1003-1005, ISSN 0892-6638

- Gasque, P., Chan, P., Fontaine, M., Ischenko, A., Lamacz, M., Gotze, O. and Morgan, B. P. (1995). Identification and characterization of the complement C5a anaphylatoxin receptor on human astrocytes. *Journal of Immunology*, Vol.155, No.10, (November 1995), pp. 4882-4889, ISSN 0022-1767
- Gasque, P., Singhrao, S. K., Neal, J. W., Gotze, O. and Morgan, B. P. (1997). Expression of the receptor for complement C5a (CD88) is up-regulated on reactive astrocytes, microglia, and endothelial cells in the inflamed human central nervous system. *The American Journal of Pathology*, Vol.150, No.1, (January 1997), pp. 31-41, ISSN 0002-9440
- Gasque, P., Singhrao, S. K., Neal, J. W., Wang, P., Sayah, S., Fontaine, M. and Morgan, B. P. (1998). The receptor for complement anaphylatoxin C3a is expressed by myeloid cells and nonmyeloid cells in inflamed human central nervous system: analysis in multiple sclerosis and bacterial meningitis. *Journal of Immunology*, Vol.160, No.7, (April 1998), pp. 3543-3554, ISSN 0022-1767
- Gavrilyuk, V., Kalinin, S., Hilbush, B. S., Middlecamp, A., McGuire, S., Pelligrino, D., Weinberg, G. and Feinstein, D. L. (2005). Identification of complement 5a-like receptor (C5L2) from astrocytes: characterization of anti-inflammatory properties. *Journal of Neurochemistry*, Vol.92, No.5, (March 2005), pp. 1140-1149, ISSN 0022-3402
- Gerard, C., Bao, L., Orozco, O., Pearson, M., Kunz, D. and Gerard, N. P. (1992). Structural diversity in the extracellular faces of peptidergic G-protein-coupled receptors. Molecular cloning of the mouse C5a anaphylatoxin receptor. *Journal of Immunology*, Vol.149, No.8, (October 1992), pp. 2600-2606, ISSN 0022-1767
- Gerard, N. P. and Gerard, C. (1991). The chemotactic receptor for human C5a anaphylatoxin. *Nature*, Vol.349, No.6310, (February 1991), pp. 614-617, ISSN 0028-0836
- Gerard, N. P., Bao, L., Xiao-Ping, H., Eddy, R. L., Jr., Shows, T. B. and Gerard, C. (1993). Human chemotaxis receptor genes cluster at 19q13.3-13.4. Characterization of the human C5a receptor gene. *Biochemistry*, Vol.32, No.5, (February 1993), pp. 1243-1250, ISSN
- Gerard, N. P., Lu, B., Liu, P., Craig, S., Fujiwara, Y., Okinaga, S. and Gerard, C. (2005). An anti-inflammatory function for the complement anaphylatoxin C5a-binding protein, C5L2. *Journal of Biological Chemistry*, Vol.280, No.48, (December 2005), pp. 39677-39680, ISSN 0021-9258
- Gerber, B. O., Meng, E. C., Dotsch, V., Baranski, T. J. and Bourne, H. R. (2001). An activation switch in the ligand binding pocket of the C5a receptor. *Journal of Biological Chemistry*, Vol.276, No.5, (February 2001), pp. 3394-3400, ISSN 0021-9258
- GeurtsvanKessel, C. H. and Lambrecht, B. N. (2008). Division of labor between dendritic cell subsets of the lung. *Mucosal Immunology*, Vol.1, No.6, (November 2008), pp. 442-450, ISSN 1933-0219
- Giannini, E., Brouchon, L. and Boulay, F. (1995). Identification of the major phosphorylation sites in human C5a anaphylatoxin receptor in vivo. *Journal of Biological Chemistry*, Vol.270, No.32, (August 1995), pp. 19166-19172, ISSN 0021-9258
- Gonczi, Z., Varga, L., Hidvegi, T., Schmidt, B., Panya, A., Kokai, M. and Fust, G. (1997). The severity of clinical symptoms in ragweed-allergic patients is related to the extent of ragweed-induced complement activation in their sera. *Allergy*, Vol.52, No.11, (November 1997), pp. 1110-1114, ISSN 0105-4538
- Goya, T., Morisaki, T. and Torisu, M. (1994). Immunologic assessment of host defense impairment in patients with septic multiple organ failure: relationship between

- complement activation and changes in neutrophil function. *Surgery*, Vol.115, No.2, (February 1994), pp. 145-155, ISSN 0039-6060
- Groeneveld, A. B., Tacx, A. N., Bossink, A. W., van Mierlo, G. J. and Hack, C. E. (2003). Circulating inflammatory mediators predict shock and mortality in febrile patients with microbial infection. *Clinical Immunology*, Vol.106, No.2, (February 2003), pp. 106-115, ISSN 1521-6616
- Gueler, F., Rong, S., Gwinner, W., Mengel, M., Brocker, V., Schon, S., Greten, T. F., Hawlisch, H., Polakowski, T., Schnatbaum, K., Menne, J., Haller, H. and Shushakova, N. (2008). Complement 5a receptor inhibition improves renal allograft survival. *Journal of the American Society of Nephrology*, Vol.19, No.12, (December 2008), pp. 2302-2312, ISSN 1046-6673
- Guo, Q., Subramanian, H., Gupta, K. and Ali, H. (2011). Regulation of c3a receptor signaling in human mast cells by g protein coupled receptor kinases. *PLoS One*, Vol.6, No.7, (July 2011), pp. e22559, ISSN 1932-6203
- Guo, R. F., Huber-Lang, M., Wang, X., Sarma, V., Padgaonkar, V. A., Craig, R. A., Riedemann, N. C., McClintock, S. D., Hlaing, T., Shi, M. M. and Ward, P. A. (2000). Protective effects of anti-C5a in sepsis-induced thymocyte apoptosis. *The Journal of Clinical Investigation*, Vol.106, No.10, (November 2000), pp. 1271-1280, ISSN 0021-9738
- Guo, R. F., Riedemann, N. C., Laudes, I. J., Sarma, V. J., Kunkel, R. G., Dilley, K. A., Paulauskis, J. D. and Ward, P. A. (2002). Altered neutrophil trafficking during sepsis. *Journal of Immunology*, Vol.169, No.1, (July 2002), pp. 307-314, ISSN 0022-1767
- Guo, R. F., Riedemann, N. C., Bernacki, K. D., Sarma, V. J., Laudes, I. J., Reuben, J. S., Younkin, E. M., Neff, T. A., Paulauskis, J. D., Zetoune, F. S. and Ward, P. A. (2003). Neutrophil C5a receptor and the outcome in a rat model of sepsis. *Faseb Journal*, Vol.17, No.13, (October 2003), pp. 1889-1891, ISSN 0892-6638
- Gutzmer, R., Lisewski, M., Zwirner, J., Mommert, S., Diesel, C., Wittmann, M., Kapp, A. and Werfel, T. (2004). Human monocyte-derived dendritic cells are chemoattracted to C3a after up-regulation of the C3a receptor with interferons. *Immunology*, Vol.111, No.4, (April 2004), pp. 435-443, ISSN 1365-2567
- Hack, C. E., Nuijens, J. H., Felt-Bersma, R. J., Schreuder, W. O., Eerenberg-Belmer, A. J., Paardekooper, J., Bronsveld, W. and Thijs, L. G. (1989). Elevated plasma levels of the anaphylatoxins C3a and C4a are associated with a fatal outcome in sepsis. *The American Journal of Medicine*, Vol.86, No.1, (January 1989), pp. 20-26, ISSN 0002-9343
- Han, G., Geng, S., Li, Y., Chen, G., Wang, R., Li, X., Ma, Y., Shen, B. and Li, Y. (2011) gammadeltaT-cell function in sepsis is modulated by C5a receptor signalling. *Immunology*, Vol.133, No.3, pp. 340-349, ISSN 1365-2567
- Hart, M. L., Walsh, M. C. and Stahl, G. L. (2004). Initiation of complement activation following oxidative stress. In vitro and in vivo observations. *Molecular Immunology*, Vol.41, No.2-3, (June 2004), pp. 165-171, ISSN 0161-5890
- Hartmann, K., Henz, B. M., Kruger-Krasagakes, S., Kohl, J., Burger, R., Guhl, S., Haase, I., Lippert, U. and Zuberbier, T. (1997). C3a and C5a stimulate chemotaxis of human mast cells. *Blood*, Vol.89, No.8, (April 1997), pp. 2863-2870, ISSN 0006-4971
- Hasegawa, K., Tamari, M., Shao, C., Shimizu, M., Takahashi, N., Mao, X. Q., Yamasaki, A., Kamada, F., Doi, S., Fujiwara, H., Miyatake, A., Fujita, K., Tamura, G., Matsubara, Y., Shirakawa, T. and Suzuki, Y. (2004). Variations in the C3, C3a receptor, and C5

- genes affect susceptibility to bronchial asthma. *Human Genetics*, Vol.115, No.4, (September 2004), pp. 295-301, ISSN 0340-6717
- Hecke, F., Schmidt, U., Kola, A., Bautsch, W., Klos, A. and Kohl, J. (1997). Circulating complement proteins in multiple trauma patients--correlation with injury severity, development of sepsis, and outcome. *Critical Care Medicine*, Vol.25, No.12, (December 1997), pp. 2015-2024, ISSN 0090-3493
- Hellings, P. W., Kasran, A., Liu, Z., Vandekerckhove, P., Wuyts, A., Overbergh, L., Mathieu, C. and Ceuppens, J. L. (2003). Interleukin-17 orchestrates the granulocyte influx into airways after allergen inhalation in a mouse model of allergic asthma. *American Journal of Respiratory Cell and Molecular Biology*, Vol.28, No.1, (January 2003), pp. 42-50, ISSN 1044-1549
- Hidvegi, T., Schmidt, B., Varga, L., Dervaderics, M., Lantos, A., Gonczi, Z., Barok, J., Otos, M., Kirschfink, M., Spath, P. and et al. (1995). In vitro complement activation by ragweed allergen extract in the sera of ragweed allergic and non-allergic persons. *Immunology Letters*, Vol.48, No.1, (November 1995), pp. 65-71, ISSN 0165-2478
- Hill, J. H. and Ward, P. A. (1971). The phlogistic role of C3 leukotactic fragments in myocardial infarcts of rats. *The Journal of Experimental Medicine*, Vol.133, No.4, (April 1971), pp. 885-900, ISSN 0022-1007
- Hollmann, T. J., Haviland, D. L., Kildsgaard, J., Watts, K. and Wetsel, R. A. (1998). Cloning, expression, sequence determination, and chromosome localization of the mouse complement C3a anaphylatoxin receptor gene. *Molecular Immunology*, Vol.35, No.3, (February 1998), pp. 137-148, ISSN 0161-5890
- Hopken, U. E., Lu, B., Gerard, N. P. and Gerard, C. (1996). The C5a chemoattractant receptor mediates mucosal defence to infection. *Nature*, Vol.383, No.6595, (September 1996), pp. 86-89, ISSN 0028-0836
- Huang, R., Lian, J. P., Robinson, D. and Badwey, J. A. (1998). Neutrophils stimulated with a variety of chemoattractants exhibit rapid activation of p21-activated kinases (Paks): separate signals are required for activation and inactivation of paks. *Molecular and Cell Biology*, Vol.18, No.12, (December 1998), pp. 7130-7138, ISSN 1098-5549
- Huber-Lang, M., Sarma, J.V., McGuire, S.R., Lu, K.T., Guo, R.F., Padgaonkar, V.A., Younkin, E.M., Laudes, I.J., Riedmann, N.C., Younger, J.G. and Ward, P.A. (2001). Protective effects of anti-C5a peptide antibodies in experimental sepsis. *Faseb Journal*, Vol.15, No.3, (January 2001), pp. 568-570, ISSN 0892-6638
- Huber-Lang, M. S., Younkin, E. M., Sarma, J. V., McGuire, S. R., Lu, K. T., Guo, R. F., Padgaonkar, V. A., Curnutte, J. T., Erickson, R. and Ward, P. A. (2002). Complement-induced impairment of innate immunity during sepsis. *Journal of Immunology*, Vol.169, No.6, (September 2002), pp. 3223-3231, ISSN 0022-1767
- Huber-Lang, M. S., Riedeman, N. C., Sarma, J. V., Younkin, E. M., McGuire, S. R., Laudes, I. J., Lu, K. T., Guo, R. F., Neff, T. A., Padgaonkar, V. A., Lambris, J. D., Spruce, L., Mastellos, D., Zetoune, F. S. and Ward, P. A. (2002). Protection of innate immunity by C5aR antagonist in septic mice. *Faseb Journal*, Vol.16, No.12, (October 2002), pp. 1567-1574, ISSN 0892-6638
- Huber-Lang, M., Younkin, E. M., Sarma, J. V., Riedemann, N., McGuire, S. R., Lu, K. T., Kunkel, R., Younger, J. G., Zetoune, F. S. and Ward, P. A. (2002). Generation of C5a by phagocytic cells. *American Journal of Pathology*, Vol.161, No.5, (November 2002), pp. 1849-1859, ISSN 0002-9440

- Huber-Lang, M. S., Sarma, J. V., McGuire, S. R., Lu, K. T., Padgaonkar, V. A., Younkin, E. M., Guo, R. F., Weber, C. H., Zuiderweg, E. R., Zetoune, F. S. and Ward, P. A. (2003). Structure-function relationships of human C5a and C5aR. *Journal of Immunology*, Vol.170, No.12, (June 2003), pp. 6115-6124, ISSN 0022-1767
- Hugli, T. E. (1981). The structural basis for anaphylatoxin and chemotactic functions of C3a, C4a, and C5a. *Critical Reviews in Immunology*, Vol.1, No.4, (February 1981), pp. 321-366, ISSN 1040-8401
- Humbles, A. A., Lu, B., Nilsson, C. A., Lilly, C., Israel, E., Fujiwara, Y., Gerard, N. P. and Gerard, C. (2000). A role for the C3a anaphylatoxin receptor in the effector phase of asthma. *Nature*, Vol.406, No.6799, (August 2000), pp. 998-1001, ISSN 0028-0836
- Jagels, M. A., Daffern, P. J. and Hugli, T. E. (2000). C3a and C5a enhance granulocyte adhesion to endothelial and epithelial cell monolayers: epithelial and endothelial priming is required for C3a-induced eosinophil adhesion. *Immunopharmacology*, Vol.46, No.3, (March 2000), pp. 209-222, ISSN 1567-5769
- Johnson, A. R., Hugli, T. E. and Muller-Eberhard, H. J. (1975). Release of histamine from rat mast cells by the complement peptides C3a and C5a. *Immunology*, Vol.28, No.6, (June 1975), pp. 1067-1080, ISSN 1365-2567
- Johswich, K., Martin, M., Thalmann, J., Rheinheimer, C., Monk, P. N. and Klos, A. (2006). Ligand specificity of the anaphylatoxin C5L2 receptor and its regulation on myeloid and epithelial cell lines. *Journal of Biological Chemistry*, Vol.281, No.51, (December 2006), pp. 39088-39095, ISSN 0021-9258
- Jordan, J. E., Montalto, M. C. and Stahl, G. L. (2001). Inhibition of mannose-binding lectin reduces postischemic myocardial reperfusion injury. *Circulation*, Vol.104, No.12, (September 2001), pp. 1413-1418, ISSN 0009-7322
- Kalant, D., MacLaren, R., Cui, W., Samanta, R., Monk, P. N., Laporte, S. A. and Cianflone, K. (2005). C5L2 is a functional receptor for acylation-stimulating protein. *Journal of Biological Chemistry*, Vol.280, No.25, (June 2005), pp. 23936-23944, ISSN 0021-9258
- Kaplan, A. P. (2004). Chronic urticaria: pathogenesis and treatment. *The Journal of Allergy and Clinical Immunology*, Vol.114, No.3, (September 2004), pp. 465-474; quiz 475, ISSN 0091-6749
- Karp, C. L., Grupe, A., Schadt, E., Ewart, S. L., Keane-Moore, M., Cuomo, P. J., Kohl, J., Wahl, L., Kuperman, D., Germer, S., Aud, D., Peltz, G. and Wills-Karp, M. (2000). Identification of complement factor 5 as a susceptibility locus for experimental allergic asthma. *Nature Immunology*, Vol.1, No.3, (September 2000), pp. 221-226, ISSN 1529-2908
- Karp, D. R. (2005). Complement and systemic lupus erythematosus. *Current Opinions in Rheumatology*, Vol.17, No.5, (September 2005), pp. 538-542, ISSN 1040-8711
- Kew, R. R., Ghebrehiwet, B. and Janoff, A. (1987). Characterization of the third component of complement (C3) after activation by cigarette smoke. *Clinical Immunology and Immunopathology*, Vol.44, No.2, (August 1987), pp. 248-258, ISSN 0090-1229
- Khirwadkar, K., Zilow, G., Oppermann, M., Kabelitz, D. and Rother, K. (1993). Interleukin-4 augments production of the third complement component by the alveolar epithelial cell line A549. *International Archives of Allergy and Immunology*, Vol.100, No.1, pp. 35-41, ISSN 1018-2438
- Kikuchi, Y. and Kaplan, A. P. (2002). A role for C5a in augmenting IgG-dependent histamine release from basophils in chronic urticaria. *The Journal of Allergy and Clinical Immunology*, Vol.109, No.1, (January 2002), pp. 114-118, ISSN 0091-6749

- Kohl, J., Baelder, R., Lewkowich, I. P., Pandey, M. K., Hawlisch, H., Wang, L., Best, J., Herman, N. S., Sproles, A. A., Zwirner, J., Whitsett, J. A., Gerard, C., Sfyroera, G., Lambris, J. D. and Wills-Karp, M. (2006). A regulatory role for the C5a anaphylatoxin in type 2 immunity in asthma. *The Journal of Clinical Investigation*, Vol.116, No.3, (March 2006), pp. 783-796, ISSN 0021-9738
- Korosec, P., Subic, T., Adamic, K., Silar, M. and Kosnik, M. (2009). C5a-induced in vitro basophil activation in patients with chronic urticaria: a pilot study. *Wien Klin Wochenschr*, Vol.121, No.9-10, pp. 339-343, ISSN 0043-5325
- Krug, N., Tschernig, T., Erpenbeck, V. J., Hohlfeld, J. M. and Kohl, J. (2001). Complement factors C3a and C5a are increased in bronchoalveolar lavage fluid after segmental allergen provocation in subjects with asthma. *American Journal of Respiratory and Critical Care Medicine*, Vol.164, No.10 Pt 1, (November 2001), pp. 1841-1843, ISSN 1073-449X
- Kuehn, H. S. and Gilfillan, A. M. (2007). G protein-coupled receptors and the modification of FcεRI-mediated mast cell activation. *Immunology Letters*, Vol.113, No.2, (November 2007), pp. 59-69, ISSN 0165-2478
- Kupp, L. I., Kosco, M. H., Schenkein, H. A. and Tew, J. G. (1991). Chemotaxis of germinal center B cells in response to C5a. *European Journal of Immunology*, Vol.21, No.11, (November 1991), pp. 2697-2701, ISSN 0022-1767
- Kuroki, M. and O'Flaherty, J. T. (1999). Extracellular signal-regulated protein kinase (ERK)-dependent and ERK-independent pathways target STAT3 on serine-727 in human neutrophils stimulated by chemotactic factors and cytokines. *Biochemistry Journal*, Vol.341 (Pt 3), (August 1 1999), pp. 691-696, ISSN 0264-6021
- Lacy, M., Jones, J., Whittemore, S. R., Haviland, D. L., Wetsel, R. A. and Barnum, S. R. (1995). Expression of the receptors for the C5a anaphylatoxin, interleukin-8 and FMLP by human astrocytes and microglia. *Journal of Neuroimmunology*, Vol.61, No.1, (August 1995), pp. 71-78, ISSN 0165-5728
- Lajoie, S., Lewkowich, I. P., Suzuki, Y., Clark, J. R., Sproles, A. A., Dienger, K., Budelsky, A. L. and Wills-Karp, M. (2010). Complement-mediated regulation of the IL-17A axis is a central genetic determinant of the severity of experimental allergic asthma. *Nature Immunology*, Vol.11, No.10, (October 2010), pp. 928-935, ISSN 1529-2908
- Lambrecht, B. N., De Veerman, M., Coyle, A. J., Gutierrez-Ramos, J. C., Thielemans, K. and Pauwels, R. A. (2000). Myeloid dendritic cells induce Th2 responses to inhaled antigen, leading to eosinophilic airway inflammation. *The Journal of Clinical Investigation*, Vol.106, No.4, (August 2000), pp. 551-559, ISSN 0021-9738
- Lambrecht, B. N. and Hammad, H. (2009). Biology of lung dendritic cells at the origin of asthma. *Immunity*, Vol.31, No.3, (September 2009), pp. 412-424, ISSN 1074-7613
- Langkabel, P., Zwirner, J. and Oppermann, M. (1999). Ligand-induced phosphorylation of anaphylatoxin receptors C3aR and C5aR is mediated by G protein-coupled receptor kinases. *European Journal of Immunology*, Vol.29, No.9, (September 1999), pp. 3035-3046, ISSN 0014-2980
- Laudes, I. J., Chu, J. C., Sikrath, S., Huber-Lang, M., Guo, R. F., Riedemann, N., Sarma, J. V., Schmaier, A. H. and Ward, P. A. (2002). Anti-c5a ameliorates coagulation/fibrinolytic protein changes in a rat model of sepsis. *American Journal of Pathology*, Vol.160, No.5, (May 2002), pp. 1867-1875, ISSN 0002-9440
- Lee, D. K., George, S. R., Cheng, R., Nguyen, T., Liu, Y., Brown, M., Lynch, K. R. and O'Dowd, B. F. (2001). Identification of four novel human G protein-coupled

- receptors expressed in the brain. *Brain Research. Molecular Brain Research*, Vol.86, No.1-2, (January 2001), pp. 13-22
- Lee, S. H., Rhim, T., Choi, Y. S., Min, J. W., Kim, S. H., Cho, S. Y., Paik, Y. K. and Park, C. S. (2006). Complement C3a and C4a increased in plasma of patients with aspirin-induced asthma. *American Journal of Respiratory and Critical Care Medicine*, Vol.173, No.4, (February 2006), pp. 370-378, ISSN 0090-3493
- Lett-Brown, M. A. and Leonard, E. J. (1977). Histamine-induced inhibition of normal human basophil chemotaxis to C5a. *Journal of Immunology*, Vol.118, No.3, (March 1977), pp. 815-818, ISSN 0022-1767
- Lewis, A. G., Kohl, G., Ma, Q., Devarajan, P. and Kohl, J. (2008). Pharmacological targeting of C5a receptors during organ preservation improves kidney graft survival. *Clinical and Experimental Immunology*, Vol.153, No.1, (July 2008), pp. 117-126, ISSN 0009-9104
- Lewkowich, I. P., Herman, N. S., Schleifer, K. W., Dance, M. P., Chen, B. L., Dienger, K. M., Sproles, A. A., Shah, J. S., Kohl, J., Belkaid, Y. and Wills-Karp, M. (2005). CD4+CD25+ T cells protect against experimentally induced asthma and alter pulmonary dendritic cell phenotype and function. *Journal of Experimental Medicine*, Vol.202, No.11, (December 2005), pp. 1549-1561, ISSN 0022-1007
- Li, K., Anderson, K. J., Peng, Q., Noble, A., Lu, B., Kelly, A. P., Wang, N., Sacks, S. H. and Zhou, W. (2008). Cyclic AMP plays a critical role in C3a-receptor-mediated regulation of dendritic cells in antigen uptake and T-cell stimulation. *Blood*, Vol.112, No.13, (December 2008), pp. 5084-5094, ISSN 0006-4971
- Link, C., Hawlisch, H., Meyer zu Vilsendorf, A., Gyleruz, S., Nagel, E. and Kohl, J. (1999). Selection of phage-displayed anti-guinea pig C5 or C5a antibodies and their application in xenotransplantation. *Molecular Immunology*, Vol.36, No.18, (December 1999), pp. 1235-1247, ISSN 0161-5890
- Lo, R. K., Wise, H. and Wong, Y. H. (2006). Prostacyclin receptor induces STAT1 and STAT3 phosphorylations in human erythroleukemia cells: a mechanism requiring PTX-insensitive G proteins, ERK and JNK. *Cell Signaling*, Vol.18, No.3, (March 2006), pp. 307-317, ISSN 0898-6568
- Lukacs, N. W., Glovsky, M. M. and Ward, P. A. (2001). Complement-dependent immune complex-induced bronchial inflammation and hyperreactivity. *American Journal of Physiology. Lung Cellular and Molecular Physiology*, Vol.280, No.3, (March 2001), pp. L512-518, ISSN 1040-0605
- Marc, M. M., Korosec, P., Kosnik, M., Kern, I., Flezar, M., Suskovic, S. and Sorli, J. (2004). Complement factors c3a, c4a, and c5a in chronic obstructive pulmonary disease and asthma. *American Journal of Respiratory Cell and Molecular Biology*, Vol.31, No.2, (August 2004), pp. 216-219, ISSN 1073-449X
- Markiewski, M. M., DeAngelis, R. A., Benencia, F., Ricklin-Lichtsteiner, S. K., Koutoulaki, A., Gerard, C., Coukos, G. and Lambris, J. D. (2008). Modulation of the antitumor immune response by complement. *Nature Immunology*, Vol.9, No.11, (November 2008), pp. 1225-1235, ISSN 1529-2908
- Maruo, K., Akaike, T., Ono, T., Okamoto, T. and Maeda, H. (1997). Generation of anaphylatoxins through proteolytic processing of C3 and C5 by house dust mite protease. *The Journal of Allergy and Clinical Immunology*, Vol.100, No.2, (August 1997), pp. 253-260, ISSN 0091-6749

- Mastellos, D., Germenis, A. E. and Lambris, J. D. (2005). Complement: an inflammatory pathway fulfilling multiple roles at the interface of innate immunity and development. *Current Drug Targets in Inflammation and Allergy*, Vol.4, No.1, (February 2005), pp. 125-127, ISSN 1871-5281
- McCarthy, K. and Henson, P. M. (1979). Induction of lysosomal enzyme secretion by alveolar macrophages in response to the purified complement fragments C5a and C5a des-arg. *Journal of Immunology*, Vol.123, No.6, (December 1979), pp. 2511-2517, ISSN 0022-1767
- McCloskey M.A., Fan Y and Luther S. (1999). Chemotaxis of rat mast cells toward adenine nucleotides. *Journal of Immunology*, Vol.163, No.2, (July 1999), pp. 970-977, ISSN 0022-1767
- McColl, B. W., Allan, S. M. and Rothwell, N. J. (2009). Systemic infection, inflammation and acute ischemic stroke. *Neuroscience*, Vol.158, No.3, (February 2009), pp. 1049-1061, ISSN 0306-4522
- McKinley, L., Alcorn, J. F., Peterson, A., Dupont, R. B., Kapadia, S., Logar, A., Henry, A., Irvin, C. G., Piganelli, J. D., Ray, A. and Kolls, J. K. (2008). TH17 cells mediate steroid-resistant airway inflammation and airway hyperresponsiveness in mice. *Journal of Immunology*, Vol.181, No.6, (September 2008), pp. 4089-4097, ISSN 0022-1767
- Melendi, G. A., Hoffman, S. J., Karron, R. A., Irusta, P. M., Laham, F. R., Humbles, A., Schofield, B., Pan, C. H., Rabold, R., Thumar, B., Thumar, A., Gerard, N. P., Mitzner, W., Barnum, S. R., Gerard, C., Kleeberger, S. R. and Polack, F. P. (2007). C5 modulates airway hyperreactivity and pulmonary eosinophilia during enhanced respiratory syncytial virus disease by decreasing C3a receptor expression. *Journal of Virology*, Vol.81, No.2, (January 2007), pp. 991-999, ISSN 0022-538X
- Mery, L. and Boulay, F. (1993). Evidence that the extracellular N-terminal domain of C5aR contains amino-acid residues crucial for C5a binding. *European Journal of Haematology*, Vol.51, No.5, (November 1993), pp. 282-287, ISSN 0902-4441
- Methner, A., Hermey, G., Schinke, B. and Hermans-Borgmeyer, I. (1997). A novel G protein-coupled receptor with homology to neuropeptide and chemoattractant receptors expressed during bone development. *Biochemical and Biophysical Research Communications*, Vol.233, No.2, (April 1997), pp. 336-342, ISSN 0006-291X
- Mizutani, N., Nabe, T. and Yoshino, S. (2009). Complement C3a regulates late asthmatic response and airway hyperresponsiveness in mice. *Journal of Immunology*, Vol.183, No.6, (September 2009), pp. 4039-4046, ISSN 0022-1767
- Mocco, J., Mack, W. J., Ducruet, A. F., Sosunov, S. A., Sughrue, M. E., Hassid, B. G., Nair, M. N., Laufer, I., Komotar, R. J., Claire, M., Holland, H., Pinsky, D. J. and Connolly, E. S., Jr. (2006). Complement component C3 mediates inflammatory injury following focal cerebral ischemia. *Circulation Research*, Vol.99, No.2, (July 2006), pp. 209-217, ISSN 0009-7330
- Monk, P. N. and Partridge, L. J. (1993). Characterization of a complement-fragment-C5a-stimulated calcium-influx mechanism in U937 monocytic cells. *Biochemistry Journal*, Vol.295 (Pt 3), (November 1993), pp. 679-684, ISSN 0264-6021
- Monk, P. N., Scola, A. M., Madala, P. and Fairlie, D. P. (2007). Function, structure and therapeutic potential of complement C5a receptors. *British Journal of Pharmacology*, Vol.152, No.4, (October 2007), pp. 429-448, ISSN 1476-5381

- Nagata, S. and Glovsky, M. M. (1987). Activation of human serum complement with allergens. I. Generation of C3a, C4a, and C5a and induction of human neutrophil aggregation. *The Journal of Allergy and Clinical Immunology*, Vol.80, No.1, (July 1987), pp. 24-32, ISSN 0091-6749
- Nakae, H., Endo, S., Inada, K., Takakuwa, T., Kasai, T. and Yoshida, M. (1994). Serum complement levels and severity of sepsis. *Research Communications in Chemical Pathology and Pharmacology*, Vol.84, No.2, (May 1994), pp. 189-195, ISSN 0034-5164
- Nakae, H., Endo, S., Inada, K. and Yoshida, M. (1996). Chronological changes in the complement system in sepsis. *Surgery Today*, Vol.26, No.4, pp. 225-229, ISSN 0941-1291
- Nakamura, T., Xi, G., Hua, Y., Schallert, T., Hoff, J. T. and Keep, R. F. (2004). Intracerebral hemorrhage in mice: model characterization and application for genetically modified mice. *Journal of Cerebral Blood Flow and Metabolism*, Vol.24, No.5, (May 2004), pp. 487-494, ISSN 0271-678X
- Nataf, S., Davoust, N., Ames, R. S. and Barnum, S. R. (1999). Human T cells express the C5a receptor and are chemoattracted to C5a. *Journal of Immunology*, Vol.162, No.7, (April 1999), pp. 4018-4023, ISSN 0022-1767
- Niederbichler, A. D., Hoesel, L. M., Westfall, M. V., Gao, H., Ipaktchi, K. R., Sun, L., Zetoune, F. S., Su, G. L., Arbabi, S., Sarma, J. V., Wang, S. C., Hemmila, M. R. and Ward, P. A. (2006). An essential role for complement C5a in the pathogenesis of septic cardiac dysfunction. *Journal of Experimental Medicine*, Vol.203, No.1, (January 2006), pp. 53-61, ISSN 0022-1007
- Nilsson, G., Johnell, M., Hammer, C. H., Tiffany, H. L., Nilsson, K., Metcalfe, D. D., Siegbahn, A. and Murphy, P. M. (1996). C3a and C5a are chemotaxins for human mast cells and act through distinct receptors via a pertussis toxin-sensitive signal transduction pathway. *Journal of Immunology*, Vol.157, No.4, (August 1996), pp. 1693-1698, ISSN 0022-1767
- Nishino, H., Czurko, A., Fukuda, A., Hashitani, T., Hida, H., Karadi, Z. and Lenard, L. (1994). Pathophysiological process after transient ischemia of the middle cerebral artery in the rat. *Brain Research Bulletin*, Vol.35, No.1, pp. 51-56, ISSN 0361-9230
- Ober, C., Cox, N. J., Abney, M., Di Rienzo, A., Lander, E. S., Changyaleket, B., Gidley, H., Kurtz, B., Lee, J., Nance, M., Pettersson, A., Prescott, J., Richardson, A., Schlenker, E., Summerhill, E., Willadsen, S. and Parry, R. (1998). Genome-wide search for asthma susceptibility loci in a founder population. The Collaborative Study on the Genetics of Asthma. *Human Molecular Genetics*, Vol.7, No.9, (September 1998), pp. 1393-1398, ISSN 0964-6906
- Ohno, M., Hirata, T., Enomoto, M., Araki, T., Ishimaru, H. and Takahashi, T. A. (2000). A putative chemoattractant receptor, C5L2, is expressed in granulocyte and immature dendritic cells, but not in mature dendritic cells. *Molecular Immunology*, Vol.37, No.8, (June 2000), pp. 407-412, ISSN 0161-5890
- Okinaga, S., Slattery, D., Humbles, A., Zsengeller, Z., Morteau, O., Kinrade, M. B., Brodbeck, R. M., Krause, J. E., Choe, H. R., Gerard, N. P. and Gerard, C. (2003). C5L2, a nonsignaling C5A binding protein. *Biochemistry*, Vol.42, No.31, (August 2003), pp. 9406-9415, ISSN 0006-2960
- Palomares, O., Yaman, G., Azkur, A. K., Akkoc, T., Akdis, M. and Akdis, C. A. (2010). Role of Treg in immune regulation of allergic diseases, *European Journal of Immunology*, Vol.40, No.5, (May 2010), pp. 1232-1240, ISSN 0014-2980

- Park, J. W., Taube, C., Joetham, A., Takeda, K., Kodama, T., Dakhama, A., McConville, G., Allen, C. B., Sfyroera, G., Shultz, L. D., Lambris, J. D., Giclas, P. C., Holers, V. M. and Gelfand, E. W. (2004). Complement activation is critical to airway hyperresponsiveness after acute ozone exposure. *American Journal of Respiratory Cell and Molecular Biology*, Vol.169, No.6, (March 2004), pp. 726-732, ISSN 1044-1549
- Park, K. W., Tofukuji, M., Metais, C., Comunale, M. E., Dai, H. B., Simons, M., Stahl, G. L., Agah, A. and Sellke, F. W. (1999). Attenuation of endothelium-dependent dilation of pig pulmonary arterioles after cardiopulmonary bypass is prevented by monoclonal antibody to complement C5a. *Anesthesia and Analgesia*, Vol.89, No.1, (July 1999), pp. 42-48, ISSN 0003-2999
- Pene, J., Chevalier, S., Preisser, L., Venereau, E., Guilleux, M. H., Ghannam, S., Moles, J. P., Danger, Y., Ravon, E., Lesaux, S., Yssel, H. and Gascan, H. (2008). Chronically inflamed human tissues are infiltrated by highly differentiated Th17 lymphocytes. *Journal of Immunology*, Vol.180, No.11, (June 2008), pp. 7423-7430, ISSN 0022-1767
- Peng, Q., Li, K., Anderson, K., Farrar, C. A., Lu, B., Smith, R. A., Sacks, S. H. and Zhou, W. (2008). Local production and activation of complement up-regulates the allostimulatory function of dendritic cells through C3a-C3aR interaction. *Blood*, Vol.111, No.4, (February 2008), pp. 2452-2461, ISSN 0006-4971
- Peng, Q., Li, K., Patel, H., Sacks, S. H. and Zhou, W. (2006). Dendritic cell synthesis of C3 is required for full T cell activation and development of a Th1 phenotype. *Journal of Immunology*, Vol.176, No.6, (March 2006), pp. 3330-3341, ISSN 0022-1767
- Peng, T., Hao, L., Madri, J. A., Su, X., Elias, J. A., Stahl, G. L., Squinto, S. and Wang, Y. (2005). Role of C5 in the development of airway inflammation, airway hyperresponsiveness, and ongoing airway response. *The Journal of Clinical Investigation*, Vol.115, No.6, (June 2005), pp. 1590-1600, ISSN 0021-9738
- Perianayagam, M. C., Balakrishnan, V. S., Pereira, B. J. and Jaber, B. L. (2004). C5a delays apoptosis of human neutrophils via an extracellular signal-regulated kinase and Bad-mediated signalling pathway. *European Journal of Clinical Investigation*, Vol.34, No.1, (January 2004), pp. 50-56, ISSN 0021-9738
- Perianayagam, M. C., Madias, N. E., Pereira, B. J. and Jaber, B. L. (2006). CREB transcription factor modulates Bcl2 transcription in response to C5a in HL-60-derived neutrophils. *European Journal of Clinical Investigation*, Vol.36, No.5, (May 2006), pp. 353-361, ISSN 0014-2972
- Pieters, W. R., Houben, L. A., Koenderman, L. and Raaijmakers, J. A. (1995). C5a-induced migration of human monocytes is primed by dexamethasone. *American Journal of Respiratory Cell and Molecular Biology*, Vol.12, No.6, (June 1995), pp. 691-696, ISSN 1044-1549
- Pratt, J. R., Basheer, S. A. and Sacks, S. H. (2002). Local synthesis of complement component C3 regulates acute renal transplant rejection. *Nature Medicine*, Vol.8, No.6, (June 2002), pp. 582-587, ISSN 1078-8956
- Premont, R. T. and Gainetdinov, R. R. (2007). Physiological roles of G protein-coupled receptor kinases and arrestins. *Annual Review of Physiology*, Vol.69, pp. 511-534, ISSN 0066-4278
- Proctor, L. M., Arumugam, T. V., Shiels, I., Reid, R. C., Fairlie, D. P. and Taylor, S. M. (2004). Comparative anti-inflammatory activities of antagonists to C3a and C5a receptors in a rat model of intestinal ischaemia/reperfusion injury. *British Journal of Pharmacology*, Vol.142, No.4, (June 2004), pp. 756-764, ISSN 1476-5381

- Pundir P and Kulka M. (2010). The role of G protein-coupled receptors in mast cell activation by antimicrobial peptides: is there a connection? *Immunology & Cell Biology*, Vol.88, No.6, (August 2010), pp. 632-640, ISSN 0818-9641
- Rabiet, M. J., Huet, E. and Boulay, F. (2007). The N-formyl peptide receptors and the anaphylatoxin C5a receptors: an overview. *Biochimie*, Vol.89, No.9, (September 2007), pp. 1089-1106, ISSN 0300-9084
- Richardson, R. M., Pridgen, B. C., Haribabu, B., Ali, H. and Snyderman, R. (1998). Differential cross-regulation of the human chemokine receptors CXCR1 and CXCR2. Evidence for time-dependent signal generation. *Journal of Biological Chemistry*, Vol.273, No.37, (September 1998), pp. 23830-23836, ISSN 0021-9258
- Ricklin, D., Hajishengallis, G., Yang, K. and Lambris, J. D. (2010). Complement: a key system for immune surveillance and homeostasis. *Nature Immunology*, Vol.11, No.9, (September 2010), pp. 785-797, ISSN 1529-2908
- Riedemann, N. C., Guo, R. F., Laudes, I. J., Keller, K., Sarma, V. J., Padgaonkar, V., Zetoune, F. S. and Ward, P. A. (2002). C5a receptor and thymocyte apoptosis in sepsis. *Faseb Journal*, Vol.16, No.8, (June 2002), pp. 887-888, ISSN 0892-6638
- Riedemann, N. C., Guo, R. F., Neff, T. A., Laudes, I. J., Keller, K. A., Sarma, V. J., Markiewski, M. M., Mastellos, D., Strey, C. W., Pierson, C. L., Lambris, J. D., Zetoune, F. S. and Ward, P. A. (2002). Increased C5a receptor expression in sepsis. *The Journal of Clinical Investigation*, Vol.110, No.1, (July 2002), pp. 101-108, ISSN 0021-9738
- Rittirsch, D., Flierl, M. A., Nadeau, B. A., Day, D. E., Huber-Lang, M., Mackay, C. R., Zetoune, F. S., Gerard, N. P., Cianflone, K., Kohl, J., Gerard, C., Sarma, J. V. and Ward, P. A. (2008). Functional roles for C5a receptors in sepsis. *Nature Medicine*, Vol.14, No.5, (May 2008), pp. 551-557, ISSN 1078-8956
- Rousseau, S., Dolado, I., Beardmore, V., Shpiro, N., Marquez, R., Nebreda, A. R., Arthur, J. S., Case, L. M., Tessier-Lavigne, M., Gaestel, M., Cuenda, A. and Cohen, P. (2006). CXCL12 and C5a trigger cell migration via a PAK1/2-p38alpha MAPK-MAPKAP-K2-HSP27 pathway. *Cell Signaling*, Vol.18, No.11, (November 2006), pp. 1897-1905, ISSN 0898-6568
- Rus, H., Cudrici, C. and Niculescu, F. (2005). C5b-9 complement complex in autoimmune demyelination and multiple sclerosis: dual role in neuroinflammation and neuroprotection. *Annals of Medicine*, Vol.37, No.2, pp. 97-104, ISSN 0785-3890
- Rynkowski, M. A., Kim, G. H., Garrett, M. C., Zacharia, B. E., Otten, M. L., Sosunov, S. A., Komotar, R. J., Hassid, B. G., Ducruet, A. F., Lambris, J. D. and Connolly, E. S. (2009). C3a receptor antagonist attenuates brain injury after intracerebral hemorrhage. *Journal of Cerebral Blood Flow and Metabolism*, Vol.29, No.1, (January 2009), pp. 98-107, ISSN 0271-678X
- Sachse, C., Wolterink, G. and Pallua, N. (2000). Neutrophil intracellular pH and phagocytosis after thermal trauma. *Clinica Chimica Acta*, Vol.295, No.1-2, (May 2000), pp. 13-26, ISSN 0009-8981
- Samson, M., Edinger, A. L., Stordeur, P., Rucker, J., Verhasselt, V., Sharron, M., Govaerts, C., Mollereau, C., Vassart, G., Doms, R. W. and Parmentier, M. (1998). ChemR23, a putative chemoattractant receptor, is expressed in monocyte-derived dendritic cells and macrophages and is a coreceptor for SIV and some primary HIV-1 strains. *European Journal of Immunology*, Vol.28, No.5, (May 1998), pp. 1689-1700, ISSN 0014-2980

- Santini, F., Gaidarov, I. and Keen, J. H. (2002). G protein-coupled receptor/arrestin3 modulation of the endocytic machinery. *Journal of Cell Biology*, Vol.156, No.4, (February 2002), pp. 665-676, ISSN 0021-9525
- Sarma, J. V. and Ward, P. A. (2011). The complement system. *Cell and Tissue Research*, Vol.343, No.1, (January 2011), pp. 227-235, ISSN 0302-766X
- Sayah, S., Patte, C., Gasque, P., Chan, P., Ischenko, A., Vaudry, H. and Fontaine, M. (1997). Characterization of rat C5a anaphylatoxin receptor (C5aR): cloning of rat C5aR cDNA and study of C5aR expression by rat astrocytes. *Brain Research. Molecular Brain Research*, Vol.48, No.2, (September 1997), pp. 215-222
- Schindler, R., Gelfand, J. A. and Dinarello, C. A. (1990). Recombinant C5a stimulates transcription rather than translation of interleukin-1 (IL-1) and tumor necrosis factor: translational signal provided by lipopolysaccharide or IL-1 itself. *Blood*, Vol.76, No.8, (October 1990), pp. 1631-1638, ISSN 0006-4971
- Schraufstatter, I. U., Trieu, K., Sikora, L., Sriramaraio, P. and DiScipio, R. (2002). Complement c3a and c5a induce different signal transduction cascades in endothelial cells. *Journal of Immunology*, Vol.169, No.4, (August 2002), pp. 2102-2110, ISSN 0022-1767
- Scola, A. M., Johswich, K. O., Morgan, B. P., Klos, A. and Monk, P. N. (2009). The human complement fragment receptor, C5L2, is a recycling decoy receptor. *Molecular Immunology*, Vol.46, No.6, (March 2009), pp. 1149-1162, ISSN 0161-5890
- Scott, M. G., Benmerah, A., Muntaner, O. and Marullo, S. (2002). Recruitment of activated G protein-coupled receptors to pre-existing clathrin-coated pits in living cells. *Journal of Biological Chemistry*, Vol.277, No.5, (February 2002), pp. 3552-3559, ISSN 0021-9258
- Selberg, O., Hecker, H., Martin, M., Klos, A., Bautsch, W. and Kohl, J. (2000). Discrimination of sepsis and systemic inflammatory response syndrome by determination of circulating plasma concentrations of procalcitonin, protein complement 3a, and interleukin-6. *Critical Care Medicine*, Vol.28, No.8, (August 2000), pp. 2793-2798, ISSN 0090-3493
- Settmacher, B., Bock, D., Saad, H., Gartner, S., Rheinheimer, C., Kohl, J., Bautsch, W. and Klos, A. (1999). Modulation of C3a activity: internalization of the human C3a receptor and its inhibition by C5a. *Journal of Immunology*, Vol.162, No.12, (June 1999), pp. 7409-7416, ISSN 0022-1767
- Shima, M. and Adachi, M. (1996). Effects of environmental tobacco smoke on serum levels of acute phase proteins in schoolchildren. *Preventive Medicine*, Vol.25, No.5, (September-October 1996), pp. 617-624, ISSN 0091-7435
- Showell, H. J., Glovsky, M. M. and Ward, P. A. (1982). C3a-induced lysosomal enzyme secretion from human neutrophils: lack of inhibition by f met-leu-phe antagonists and inhibition by arachidonic acid antagonists. *International Archives of Allergy and Applied Immunology*, Vol.67, No.3, pp. 227-232, ISSN 0020-5915
- Skokowa, J., Ali, S. R., Felda, O., Kumar, V., Konrad, S., Shushakova, N., Schmidt, R. E., Piekorz, R. P., Nurnberg, B., Spicher, K., Birnbaumer, L., Zwirner, J., Claassens, J. W., Verbeek, J. S., van Rooijen, N., Kohl, J. and Gessner, J. E. (2005). Macrophages induce the inflammatory response in the pulmonary Arthus reaction through G alpha i2 activation that controls C5aR and Fc receptor cooperation. *Journal of Immunology*, Vol.174, No.5, (Mach 2005), pp. 3041-3050, ISSN 0022-1767

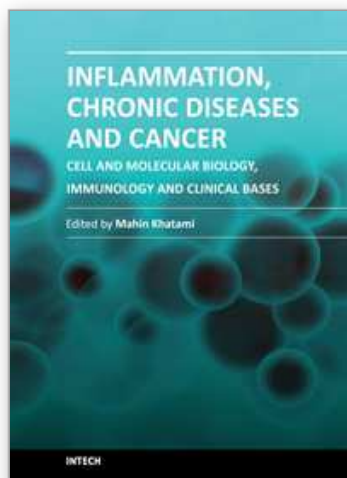
- Smith, J. K., Chi, D. S., Krish, G., Reynolds, S. and Cambron, G. (1990). Effect of exercise on complement activity. *Annals of Allergy*, Vol.65, No.4, (October 1990), pp. 304-310, ISSN 0003-4738
- Solomkin, J. S., Jenkins, M. K., Nelson, R. D., Chenoweth, D. and Simmons, R. L. (1981). Neutrophil dysfunction in sepsis. II. Evidence for the role of complement activation products in cellular deactivation. *Surgery*, Vol.90, No.2, (August 1981), pp. 319-327, ISSN 0039-6060
- Solomkin, J. S., Cotta, L. A., Satoh, P. S., Hurst, J. M. and Nelson, R. D. (1985). Complement activation and clearance in acute illness and injury: evidence for C5a as a cell-directed mediator of the adult respiratory distress syndrome in man. *Surgery*, Vol.97, No.6, (June 1985), pp. 668-678, ISSN 0039-6060
- Soruri, A., Kiafard, Z., Dettmer, C., Riggert, J., Kohl, J. and Zwirner, J. (2003). IL-4 down-regulates anaphylatoxin receptors in monocytes and dendritic cells and impairs anaphylatoxin-induced migration in vivo. *Journal of Immunology*, Vol.170, No.6, (March 2003), pp. 3306-3314, ISSN 0022-1767
- Speyer, C. L., Gao, H., Rancilio, N. J., Neff, T. A., Huffnagle, G. B., Sarma, J. V. and Ward, P. A. (2004). Novel chemokine responsiveness and mobilization of neutrophils during sepsis. *American Journal of Pathology*, Vol.165, No.6, (December 2004), pp. 2187-2196, ISSN 0002-9440
- Stevens, J. H., O'Hanley, P., Shapiro, J. M., Mihm, F. G., Satoh, P. S., Collins, J. A. and Raffin, T. A. (1986). Effects of anti-C5a antibodies on the adult respiratory distress syndrome in septic primates. *The Journal of Clinical Investigation*, Vol.77, No.6, (June 1986), pp. 1812-1816, ISSN 0021-9738
- Stimler, N. P., Bloor, C. M. and Hugli, T. E. (1983). C3a-induced contraction of guinea pig lung parenchyma: role of cyclooxygenase metabolites. *Immunopharmacology*, Vol.5, No.3, (February 1983), pp. 251-257, ISSN 1567-5769
- Subramanian, H., Kashem, S. W., Collington, S. J., Qu, H., Lambris, J. D. and Ali, H. (2011). PMX-53 as a dual CD88 antagonist and an agonist for Mas-related gene 2 (MrgX2) in human mast cells. *Molecular Pharmacology*, Vol.79, No.6, (June 2011), pp. 1005-1013, ISSN 0026-895X
- Szeplaki, G., Szegedi, R., Hirschberg, K., Gombos, T., Varga, L., Karadi, I., Entz, L., Szeplaki, Z., Garred, P., Prohaszka, Z. and Fust, G. (2009). Strong complement activation after acute ischemic stroke is associated with unfavorable outcomes. *Atherosclerosis*, Vol.204, No.1, (May 2009), pp. 315-320, ISSN 0021-9150
- Takafuji, S., Tadokoro, K., Ito, K. and Dahinden, C. A. (1994). Degranulation from human eosinophils stimulated with C3a and C5a. *International Archives of Allergy and Immunology*, Vol.104 Suppl 1, No.1, pp. 27-29, ISSN 1018-2438
- Tardif, M., Brouchon, L., Rabinet, M. J. and Boulay, F. (2003). Direct binding of a fragment of the Wiskott-Aldrich syndrome protein to the C-terminal end of the anaphylatoxin C5a receptor. *Biochemistry Journal*, Vol.372, No.Pt 2, (June 2003), pp. 453-463, ISSN 0264-6021
- Taube, C., Thurman, J. M., Takeda, K., Joetham, A., Miyahara, N., Carroll, M. C., Dakhama, A., Giclas, P. C., Holers, V. M. and Gelfand, E. W. (2006). Factor B of the alternative complement pathway regulates development of airway hyperresponsiveness and inflammation. *Proceedings of the National Academy of Science of the United States of America*, Vol.103, No.21, (May 2006), pp. 8084-8089, ISSN 0027-8424

- Thurman, J. M., Ljubanovic, D., Edelstein, C. L., Gilkeson, G. S. and Holers, V. M. (2003). Lack of a functional alternative complement pathway ameliorates ischemic acute renal failure in mice. *Journal of Immunology*, Vol.170, No.3, (February 2003), pp. 1517-1523, ISSN 0022-1767
- Tsang, S. H., Burns, M. E., Calvert, P. D., Gouras, P., Baylor, D. A., Goff, S. P. and Arshavsky, V. Y. (1998). Role for the target enzyme in deactivation of photoreceptor G protein in vivo. *Science*, Vol.282, No.5386, (October 1998), pp. 117-121, ISSN 0036-8705
- Urra, X., Cervera, A., Villamor, N., Planas, A. M. and Chamorro, A. (2009). Harms and benefits of lymphocyte subpopulations in patients with acute stroke. *Neuroscience*, Vol.158, No.3, (Feb 6 2009), pp. 1174-1183, ISSN 0306-4522
- Utoh, J., Utsunomiya, T., Imamura, T., Katsuya, H., Miyauchi, Y. and Kambara, T. (1989). Complement activation and neutrophil dysfunction in burned patients with sepsis—a study of two cases. *The Japanese Journal of Surgery*, Vol.19, No.4, (July 1989), pp. 462-467, ISSN 0047-1909
- Van Beek, J., Bernaudin, M., Petit, E., Gasque, P., Nouvelot, A., MacKenzie, E. T. and Fontaine, M. (2000). Expression of receptors for complement anaphylatoxins C3a and C5a following permanent focal cerebral ischemia in the mouse. *Experimental Neurology*, Vol.161, No.1, (January 2000), pp. 373-382, ISSN 0014-4886
- Vasthare, U. S., Barone, F. C., Sarau, H. M., Rosenwasser, R. H., DiMartino, M., Young, W. F. and Tuma, R. F. (1998). Complement depletion improves neurological function in cerebral ischemia. *Brain Research Bulletin*, Vol.45, No.4, pp. 413-419, ISSN 0361-9230
- Venkatesha, R. T., Berla Thangam, E., Zaidi, A. K. and Ali, H. (2005). Distinct regulation of C3a-induced MCP-1/CCL2 and RANTES/CCL5 production in human mast cells by extracellular signal regulated kinase and PI3 kinase. *Molecular Immunology*, Vol.42, No.5, (March 2005), pp. 581-587, ISSN 0161-5890
- Vibhuti, A., Gupta, K., Subramanian, H., Guo, Q. and Ali, H. (2011). Distinct and shared roles of beta-arrestin-1 and beta-arrestin-2 on the regulation of C3a receptor signaling in human mast cells. *PLoS One*, Vol.6, No.5, (May 2011), pp. e19585, ISSN 1932-6203
- Vieyra, M., Leisman, S., Raedler, H., Kwan, W. H., Yang, M., Strainic, M. G., Medof, M. E. and Heeger, P. S. (2011). Complement regulates CD4 T-cell help to CD8 T cells required for murine allograft rejection. *American Journal of Pathology*, Vol.179, No.2, (August 2011), pp. 766-774, ISSN 0002-9440
- Wagner, E. and Frank, M. M. (2010). Therapeutic potential of complement modulation. *Nature Reviews Drug Discovery*, Vol.9, No.1, (January 2010), pp. 43-56, ISSN 1474-1776
- Wagner, M. H., Sonntag, J., Strauss, E. and Obladen, M. (1999). Complement and contact activation related to surfactant response in respiratory distress syndrome. *Pediatric Research*, Vol.45, No.1, (January 1999), pp. 14-18, ISSN 0031-3998
- Wakashin, H., Hirose, K., Maezawa, Y., Kagami, S., Suto, A., Watanabe, N., Saito, Y., Hatano, M., Tokuhisa, T., Iwakura, Y., Puccetti, P., Iwamoto, I. and Nakajima, H. (2008). IL-23 and Th17 cells enhance Th2-cell-mediated eosinophilic airway inflammation in mice. *American Journal of Respiratory Cell and Molecular Biology*, Vol.178, No.10, (November 2008), pp. 1023-1032, ISSN 1044-1549
- Walters, D. M., Breyse, P. N., Schofield, B. and Wills-Karp, M. (2002). Complement factor 3 mediates particulate matter-induced airway hyperresponsiveness. *American Journal*

- of Respiratory Cell and Molecular Biology*, Vol.27, No.4, (October 2002), pp. 413-418, ISSN 1044-1549
- Ward, P.A. (2004). The dark side of C5a in sepsis. *Nature Reviews Immunology*, Vol.4, No.2, (February 2004), pp. 133-142, ISSN 1474-1733
- Weaver, D. J., Jr., Reis, E. S., Pandey, M. K., Kohl, G., Harris, N., Gerard, C. and Kohl, J. (2010). C5a receptor-deficient dendritic cells promote induction of Treg and Th17 cells. *European Journal of Immunology*, Vol.40, No.3, (March 2010), pp. 710-721, ISSN 0022-1767
- Webster, R. O., Zanolari, B. and Henson, P. M. (1980). Neutrophil chemotaxis in response to surface-bound C5A. *Experimental Cell Research*, Vol.129, No.1, (September 1980), pp. 55-62, ISSN 0014-4827
- Weinberg, P. F., Matthay, M. A., Webster, R. O., Roskos, K. V., Goldstein, I. M. and Murray, J. F. (1984). Biologically active products of complement and acute lung injury in patients with the sepsis syndrome. *The American Review of Respiratory Disease*, Vol.130, No.5, (November 1984), pp. 791-796, ISSN 0003-0805
- Weiser, M. R., Williams, J. P., Moore, F. D., Jr., Kobzik, L., Ma, M., Hechtman, H. B. and Carroll, M. C. (1996). Reperfusion injury of ischemic skeletal muscle is mediated by natural antibody and complement. *Journal of Experimental Medicine*, Vol.183, No.5, (May 1996), pp. 2343-2348, ISSN 0022-1007
- Willars, G. B. (2006). Mammalian RGS proteins: multifunctional regulators of cellular signalling. *Seminars in Cell and Developmental Biology*, Vol.17, No.3, (June 2006), pp. 363-376, ISSN 1084-9521
- Wills-Karp, M. (2007). Complement activation pathways: a bridge between innate and adaptive immune responses in asthma. *Proceedings of the American Thoracic Society*, Vol.4, No.3, (July 2007), pp. 247-251, ISSN 1546-3222
- Wjst, M., Fischer, G., Immervoll, T., Jung, M., Saar, K., Rueschendorf, F., Reis, A., Ulbrecht, M., Gomolka, M., Weiss, E. H., Jaeger, L., Nickel, R., Richter, K., Kjellman, N. I., Griesse, M., von Berg, A., Gappa, M., Riedel, F., Boehle, M., van Koningsbruggen, S., Schoberth, P., Szczepanski, R., Dorsch, W., Silbermann, M., Wichmann, H. E. and et al. (1999). A genome-wide search for linkage to asthma. German Asthma Genetics Group. *Genomics*, Vol.58, No.1, (May 1999), pp. 1-8, ISSN 0888-7543
- Woodruff, T. M., Arumugam, T. V., Shiels, I. A., Reid, R. C., Fairlie, D. P. and Taylor, S. M. (2004). Protective effects of a potent C5a receptor antagonist on experimental acute limb ischemia-reperfusion in rats. *The Journal of Surgical Research*, Vol.116, No.1, (January 2004), pp. 81-90, ISSN 0022-4804
- Wust, S. K., Blumenthal, M. N., Corazalla, E. O., Benson, B. A. and Dalmaso, A. P. (2006). Complement in asthma: sensitivity to activation and generation of C3a and C5a via the different complement pathways. *Translational Research: The Journal of Laboratory and Clinical Medicine*, Vol.148, No.4, (October 2006), pp. 157-163, ISSN 1931-5244
- Xu, R., Wang, R., Han, G., Wang, J., Chen, G., Wang, L., Li, X., Guo, R., Shen, B. and Li, Y. (2010). Complement C5a regulates IL-17 by affecting the crosstalk between DC and gammadelta T cells in CLP-induced sepsis. *European Journal of Immunology*, Vol.40, No.4, (April 2010), pp. 1079-1088, ISSN 0022-1767
- Yan, J., Allendorf, D. J., Li, B., Yan, R., Hansen, R. and Donev, R. (2008). The role of membrane complement regulatory proteins in cancer immunotherapy. *Advances in Experimental Medicine and Biology*, Vol.632, pp. 159-174, ISSN 0065-2598

- Yanamadala, V. and Friedlander, R. M. (2010). Complement in neuroprotection and neurodegeneration. *Trends in Molecular Medicine*, Vol.16, No.2, (February 2010), pp. 69-76, ISSN 1471-4914
- Yilmaz, G., Arumugam, T. V., Stokes, K. Y. and Granger, D. N. (2006). Role of T lymphocytes and interferon-gamma in ischemic stroke. *Circulation*, Vol.113, No.17, (May 2006), pp. 2105-2112, ISSN 0009-7322
- Zhang, H., Qin, G., Liang, G., Li, J., Barrington, R. A. and Liu, D. X. (2007). C5aR-mediated myocardial ischemia/reperfusion injury. *Biochemical and Biophysical Research Communications*, Vol.357, No.2, (June 2007), pp. 446-452, ISSN 0006-291X
- Zheng, X., Zhang, X., Feng, B., Sun, H., Suzuki, M., Ichim, T., Kubo, N., Wong, A., Min, L. R., Budohn, M. E., Garcia, B., Jevnikar, A. M. and Min, W. P. (2008). Gene silencing of complement C5a receptor using siRNA for preventing ischemia/reperfusion injury. *American of Journal Pathology*, Vol.173, No.4, (October 2008), pp. 973-980, ISSN 0002-9440
- Zhang, X., Lewkowich, I. P., Kohl, G., Clark, J. R., Wills-Karp, M. and Kohl, J. (2009). A protective role for C5a in the development of allergic asthma associated with altered levels of B7-H1 and B7-DC on plasmacytoid dendritic cells. *Journal of Immunology*, Vol.182, No.8, (April 2009), pp. 5123-5130, ISSN 0022-1767
- Zhang, X., Schmudde, I., Laumonnier, Y., Pandey, M. K., Clark, J. R., Konig, P., Gerard, N. P., Gerard, C., Wills-Karp, M. and Kohl, J. (2010). A critical role for C5L2 in the pathogenesis of experimental allergic asthma. *Journal of Immunology*, Vol.185, No.11, (December 2010), pp. 6741-6752, ISSN 0022-1767
- Zhou, W., Patel, H., Li, K., Peng, Q., Villiers, M. B. and Sacks, S. H. (2006). Macrophages from C3-deficient mice have impaired potency to stimulate alloreactive T cells. *Blood*, Vol.107, No.6, (March 2006), pp. 2461-2469, ISSN
- Zimmermann, T., Laszik, Z., Nagy, S., Kaszaki, J. and Joo, F. (1989). The role of the complement system in the pathogenesis of multiple organ failure in shock. *Progress in Clinical and Biological Research*, Vol.308, (1989), pp. 291-297, ISSN 0361-7742

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This book is a collection of excellent reviews and perspectives contributed by experts in the multidisciplinary field of basic science, clinical studies and treatment options for a wide range of acute and chronic inflammatory diseases or cancer. The goal has been to demonstrate that persistent or chronic (unresolved or subclinical) inflammation is a common denominator in the genesis, progression and manifestation of many illnesses and/or cancers, particularly during the aging process. Understanding the fundamental basis of shared and interrelated immunological features of unresolved inflammation in initiation and progression of chronic diseases or cancer are expected to hold real promises when the designs of cost-effective strategies are considered for diagnosis, prevention or treatment of a number of age-associated illnesses such as autoimmune and neurodegenerative diseases as well as many cancers.

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